TESIS DOCTORAL INTERNACIONAL

International Doctoral Thesis

EVALUACIÓN DE LA EFICIENCIA VENTILATORIA EN ATLETAS Y SU RELACIÓN CON EL RENDIMIENTO FÍSICO Y DEPORTIVO

EVALUATION OF VENTILATORY EFFICIENCY IN ATHLETES AND ITS RELATIONSHIP WITH SPORT PERFORMANCE

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Departamento de Informática y Deporte

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Eduardo Salazar Martínez

2017

A Iris,

Por ser el corazón de esta Tesis

.

A todos los que han creído en esta Tesis Doctoral:

Familia, Alfredo, José, Martin,

A mis abuelos,

A los que siempre llevaré en el recuerdo

Prof. Alfredo Santalla Hernández

Prof. José Naranjo Orellana

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Universidad Pablo de Olavide – Sevilla

EL DR. ALFREDO SANTALLA HERNÁDEZ Y EL DR. JOSÉ NARANJO ORELLANA, profesores del programa de doctorado en Ciencias de la Actividad Física y Deporte, en la Universidad Pablo de Olavide de Sevilla

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Sevilla, 10 de Julio de 2017

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“He aprendido que el mundo quiere vivir en la cima de la montaña, sin saber que la verdadera felicidad está en la forma de subir la escarpada”

Gabriel García Márquez

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TABLA CONTENIDOS / TABLE OF CONTENTS

SALAZAR-MARTÍNEZ E.

TABLA DE CONTENIDOS [Table of contents]

TABLA DE ABREVIATURAS [Table of abbreviations]

Analysis of Variance (ANOVA)
Breathing frecuency (f_R)
Carbon dioxide (VCO_2)
Carbon dioxide equivalent (VCO_2)
Carbon dioxide output (VCO_2)
Driving (VT/Ti)
Expiration time (Te)
Force vital capacity (FVC)
Heart rate (HR)
High-intensity interval training (HIIT)
Inspiratory muscle training (IMT)
Inspiratory capacity (IC)
Inspiration time (Ti)
Maximum heart rate (HRmax)
Maximum inspiratory pressure (Pimax)
Maximum oxygen uptake (VO_2max)

Maximum power (Wmax)
Mean power (Wmean)
Oxygen uptake efficiency slope (OUES)
Oxygen equivalent (EqO_2)
Oxygen uptake (VO_2)
Partial pressure of carbon dioxide (PCO_2)
Partial pressure of oxygen (PO_2)
Peak power output (PPO)
Power (W)
Respiratory exchange ratio (RER)
Tidal volume (VT)
Timming (Ti/Ttot)
Total respiration time (Ttot)
Ventilation (V_E)
Ventilatory Efficiency (V_E/VCO_2 Slope)
Vital capacity (VC)

RESUMEN / ABSTRACT

SALAZAR-MARTÍNEZ E.

RESUMEN [Abstract]

Durante la última década, la ***eficiencia ventilatoria*** ha sido objeto de estudio en el ámbito de la Medicina clínica. Su aplicación y uso en estas áreas ha sido ampliamente contrastada.

Sin embargo, la aplicación y la relación de la eficiencia ventilatoria con el rendimiento físico y deportivo es a día de hoy desconocida. No existe una evidencia científica clara que aporte información a cerca de su uso en el ámbito deportivo. De este modo, el objetivo principal de la presente Tesis Doctoral Internacional fue analizar la implicación de la eficiencia ventilatoria como factor determinante del rendimiento deportivo en sujetos sanos.

Esta Tesis Doctoral Internacional trata de abordar el mencionado objetivo desde distintas perspectivas. En primer lugar, se llevó a cabo un estudio observacional-descriptivo sobre la respuesta de la eficiencia ventilatoria en ciclistas de élite a lo largo de tres temporadas competitivas. El objetivo fue conocer la respuesta de esta variable en sujetos altamente entrenados y esclarecer si cambios en el rendimiento deportivo estaban asociados a cambios en la eficiencia ventilatoria. En segundo lugar, se describió la respuesta de la eficiencia ventilatoria en condiciones de hipoxia, así como la influencia de un programa de entrenamiento específico de los músculos inspiratorios sobre la eficiencia ventilatoria en normoxia e hipoxia. Seguidamente, se evaluó la influencia de un programa de entrenamiento interválico de alta intensidad sobre la respuesta de la eficiencia ventilatoria de sujetos entrenados. Finalmente, se llevó a cabo un profundo análisis de la eficiencia ventilatoria en deportistas, con el objetivo de conocer la influencia del nivel de entrenamiento, ergómetro utilizado, edad y composición corporal en la respuesta de la eficiencia ventilatoria.

Los principales resultados de esta Tesis Doctoral Internacional indican que: a) la eficiencia ventilatoria es un parámetro sujeto a poca variabilidad intra-sujeto; b) ni las grandes cargas de entrenamiento no controlado, ni protocolos de entrenamiento específicamente diseñados y controlados provocan cambios en la respuesta de la eficiencia ventilatoria en deportistas; c) independientemente de las características del sujeto evaluado y del instrumento usado para evaluar la eficiencia ventilatoria, ésta tiende a responder de forma similar en deportistas; d) aparentemente no existe una

relación entre la respuesta de la eficiencia ventilatoria y el rendimiento físico y deportivo.

Como conclusión general, esta Tesis Doctoral Internacional pone de manifiesto que la eficiencia ventilatoria no es un parámetro determinante del rendimiento físico y deportivo en deportistas y que el entrenamiento no modifica o influye en su respuesta.

ABSTRACT [Resumen]

During the last years, the ***ventilatory efficiency*** has been a variable widely studied in the Medical field. Many scientists have investigated the application of this paramenter in unhealthy people.

However, the relationship between ventilatory efficiency and sport performance remains unknown. There is no support which justifies the use of this variable in the sports science field. The aim of this International PhD Thesis is to analysis the influence of ventilatory efficiency on sport performance in healthy subjects.

This International PhD Thesis trys to evaluate the aim mentioned previously. Firstly, we carried out an observational study of ventilatory efficiency in world-class cyclists over three competitive seasons. We evaluated the response of this variable in trained athletes and the relationship between changes in sport performance and ventilatory efficiency. Secondly, we investigated the response of ventilatory efficiency in normoxia and hypoxia; in addition we evaluated the influence of inspiratory muscle training on ventilatory efficiency and cycling performance. Thirdly, we evaluated the influence of high-intensity interval training of ventilatory efficiency in well-trained athletes. Lastly, we carried out a wide evaluation of ventilatory efficiency in athletes. We investigated the influence of fitness level, ergometer, body mass index and age on ventilatory efficiency response.

The main findings of this International PhD Thesis show: a) there is no intra-subjects variability in the ventilatory efficiency response of sporty people; b) Neither un-controlled or controlled specific-training programmes do not influence the ventilatory efficiency response of sporty people; c) the response of ventilatory efficiency is independent of athlete’s characteristics or ergometer used during the evaluation; d) the lack of relationship between ventilatory efficiency and sport performance.

Summarising, the main outcome of this International PhD Thesis is that ventilatory efficiency is not a variable related to sport performance in healthy subjects and its response is not affected by training.

CAPÍTULO 1

MARCO TEÓRICO /

THEORETICAL CONTEXT

SALAZAR-MARTÍNEZ E.

1- MARCO TEÓRICO [Theoretical context]

1.1. EL CENTRO RESPIRATORIO (Modificado de (Hall & Guyton, 2001))

La respiración en los seres humanos esta regulada en su mayor parte por el Sistema Nervioso Central (SNC). El SNC, se encarga de ajustar la ventilación en función de las demandas del organismo con el objeto de evitar que las presiones parciales de oxígeno (PO_2) y dióxido de carbono (PCO_2) se alteren. Concretamente, el encargado de llevar a cabo esta tarea es un sistema neurogénico denominado Centro Respiratorio.

El Centro Respiratorio está compuesto por tres grupos de neuronas localizadas de forma bilateral en el bulbo raquídeo y en la protuberancia. El primer grupo de *neuronas dorsales*, se encargan de estimular la inspiración. Un segundo *grupo respiratorio ventral*, se encarga de poner en marcha la inspiración o espiración, dependiendo de cuáles sean las neuronas del grupo que se estimulen. Por último existe un tercer grupo de neuronas, denominadas *centro neumotáxico*, que se localizan dorsalmente en la parte superior de la protuberancia, las cuales ayudan a controlar la frecuencia y patrón respiratorio.

1.1.1. Neuronas Dorsales

Al grupo de neuronas dorsales llegan señales sensitivas procedentes de los quimiorreceptores periféricos, los barorreceptores y de varios tipos de receptores del pulmón.

En esta zona del centro respiratorio se produce el control del ritmo básico de la respiración. La señal nerviosa que se transmite a los músculos inspiratorios primeros como el diafragma no es una mera suma de potenciales de acción. En la respiración normal, la inspiración comienza débilmente y crece en forma de rampa durante un periodo de 2 segundos. Cesa pasados unos 3 segundos, lo que provoca que se deje de estimular el diafragma y se que se retraiga la pared torácica y los pulmones originándose de este modo la espiración.

1.1.2. Grupo Respiratorio Ventral

La función de este grupo neuronal difiere en varios aspectos del grupo respiratorio dorsal:

1. Este grupo de neuronas permanecen inactivas durante la respiración normal tranquila, por lo que no participan en la oscilación rítmica básica que controla la respiración.

2. Esta área influye al impulso respiratorio cuando se incrementa la ventilación pulmonar gracias a la propagación de las señales provenientes de la zona respiratorio dorsal.

3. La estimulación de esta zona es importante para enviar señales poderosas a los músculos abdominales durante la espiración forzada. Por lo tanto esta zona funciona como mecanismo de hiperestimulación cuando se requieren niveles elevados de ventilación pulmonar, sobre todo durante el ejercicio.

1.1.3. Centro Neumotáxico

El centro neumotáxico se encarga de transmitir señales al área inspiratoria. Cuando la señal es fuerte, la inspiración reduce su duración considerablemente (hasta 0,5 segundos) siendo el llenado pequeño. Pero cuando las señales neumotáxicas son débiles, las inspiraciones pueden llegar a durar más de 5 segundos, llenando los pulmones con un gran exceso de aire.

Por lo tanto, la función principal del centro neumotóxico consiste en limitar la duración de la inspiración. Este hecho tiene un efecto secundario sobre la frecuencia respiratoria, ya que si la duración de la fase inspiratoria se ve reducida también lo hace en consecuencia la espiratoria, viéndose alterado todo el proceso respiratorio. Una señal fuerte puede aumentar la frecuencia respiratoria hasta las 30 o 40 respiraciones por minuto, mientras que una señal débil puede reducirla a sólo 3-5 respiraciones por minuto.

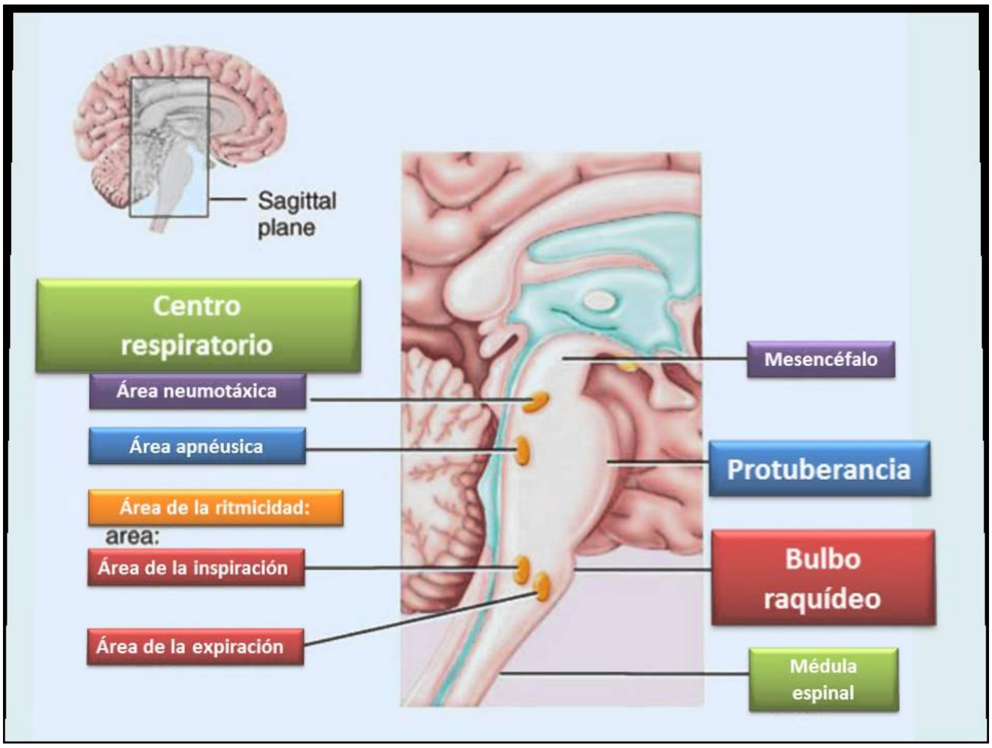


Fig. 1: Áreas del centro respiratorio central (Extraído de (Tortora & Derrickson, 2007))

1.2. REGULACIÓN DEL CENTRO RESPIRATORIO (Modificado de (Tortora & Derrickson, 2007))

La actividad del centro respiratorio puede verse modificada por ciertos factores:

1.2.1. Influencias corticales sobre la respiración

Dado que la corteza cerebral tiene conexiones con el centro respiratorio, es posible alterar voluntariamente el patrón de la respiración. En el caso de desearlo, podemos dejar de respirar por algún tiempo. Este hecho permite evitar que el agua o gases nocivos entre en los pulmones. Pero la capacidad de contener la respiración está limitada por el aumento del CO_2 e hidrogeniones (H^+) en el organismo. Estos estimulan el área inspiratoria provocando la respuesta respiratoria. Incluso si fuésemos capaces de contener la respiración hasta provocar un desmayo, ésta se vería reanudada en el momento en el que se pierde la consciencia.

1.2.2. Regulación de las respiración por quimiorreceptores

Ciertos estímulos químicos son capaces de modificar y modular la rapidez y la profundidad de la respiración. El sistema respiratorio funciona para mantener los niveles apropiados de CO_2 y O_2 en sangre en función de la demanda y respuesta del organismo, respondiendo a cambios en los niveles de estos gases.

Los quimiorreceptores controlan los niveles de CO_2, O_2 y H^+ en dos localizaciones y proveen aferencias al centro respiratorio. Los quimiorreceptores centrales están localizados en el bulbo raquídeo o en sus inmediaciones. Responden a cambios en la concentración de H^+ y/o en la presión parcial de CO_2 (PCO_2). Los quimiorreceptores periféricos están localizados en los cuerpos aórticos y carotídeos. Estos quimiorreceptores forman parte del sistema nervioso periférico y son sensibles a los cambios en la PO_2, H^+ y PCO_2 en sangre.

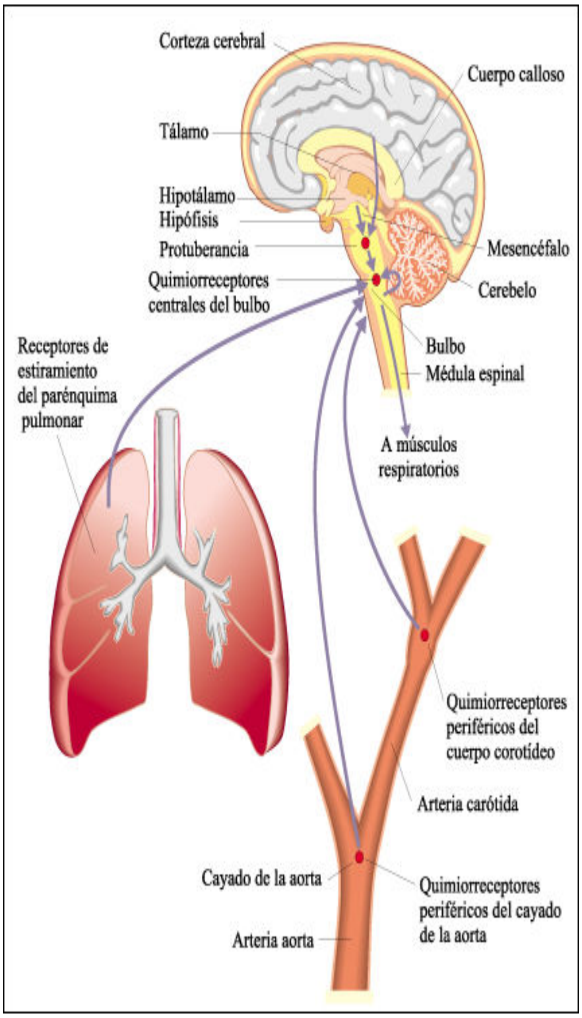


Fig. 2: Regulación de la respiración (Extraído de (Tortora & Derrickson, 2007))

Gracias a la liposolubilidad del CO_2, este traspasa fácilmente la membrana celular, donde en presencia de la anhidrasa carbónica se combina con el agua (H_2O) para formar ácido carbónico (H_2CO_3). Éste se desdobla fácilmente en H^+ y $HCO3^-$. De este modo un aumento en la PCO_2 provoca un aumento en la concrentración de H^+ con la consiguiente acidificación del pH.

Cuando realizamos ejercicio físico, se producen pequeños aumentos en la $PC0_2$, este hecho, unido al aumento en la concentración de H^+ y descenso del pH, provocan la estimulación de los quimiorreceptores centrales y periféricos que a su vez hacen que el área inspiratoria se vuelva muy activa, aumentando así de este modo la frecuencia y profundidad de la respiración con el objeto de regresar al organismo a su estado de homeostasis.

1.2.3. pH y Respuesta Vascular

Durante el ejercicio intenso tienen lugar un aumento de la concentración CO_2 en sangre que provoca un respuesta considerable de la ventilación como se ha descrito en párrafos anteriores. Pero esta no es el único proceso que se desencadena como consecuencia de este aumento en la PCO_2.

Cuando la PCO_2 es elevada, se produce un descenso del pH sanguíneo. Este descenso provoca que el diámetro de los capilares aumente (vasodilatación), con el objeto de maximizar la transferencia de glucosa y oxígeno desde el torrente sanguíneo hasta los tejidos (Gilbert, 2005)

$\downarrow$ $pH = 6{,}4 + \text{Log } [HCO_3^-] / [CO_2]$ $\uparrow$

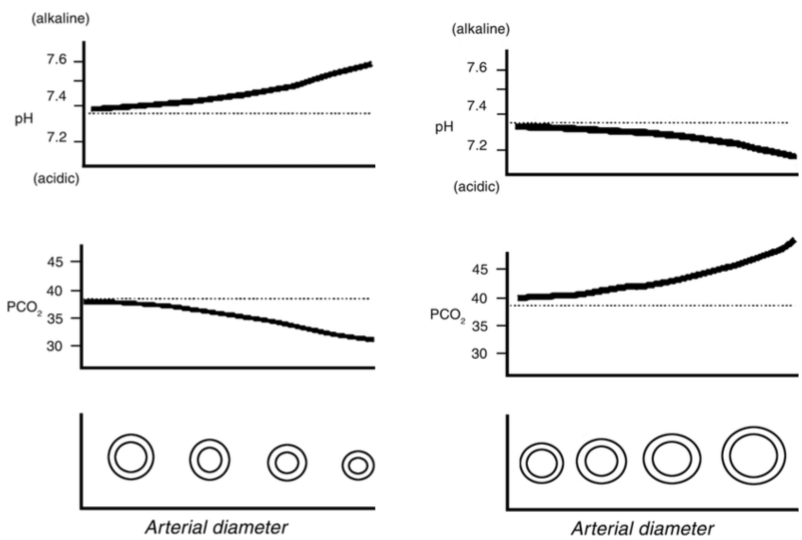


Fig.3: Relación entre la PCO_2, pH y el diámetro vascular (Extraído de (Gilbert, 2005))

De forma inversa, cuando la PCO_2 es reducida por debajo de los niveles basales, promovida por una ventilación excesiva, el pH sanguíneo se vuelve alcalino. Este hecho provoca una vasoconstricción en los capilares, inhibiendo la transferencia de nutrientes y 0_2 desde el torrente sanguíneo (Gilbert, 2005).

$$\uparrow \quad pH = 6{,}4 + \text{Log}\,[HCO_3^-] / [CO_2] \quad \downarrow$$

CAPÍTULO 2

INTRODUCCIÓN / INTRODUCTION

SALAZAR-MARTÍNEZ E.

2- INTRODUCCIÓN [Introduction]

2.1. LA EFICIENCIA VENTILATORIA

2.1.1. Concepto

Como se ha expuesto en párrafos anteriores, la producción de CO_2 y la respuesta ventilatoria se encuentran interrelacionadas profundamente. De la modificación de una nace la respuesta de la otra. Gracias al estudio de esta interrelación podemos evaluar la eficiencia ventilatoria.

De acuerdo con Sun, Hansen, Garatachea, Storer, and Wasserman (2002), el pulmón más eficiente será aquel que consiga eliminar una cantidad de CO_2 dada, con un menor nivel de ventilación. En esta misma línea Davis et al. (2006) definen la eficiencia ventilatoria como el nivel de ventilación expresado para conseguir eliminar una determinada cantidad de CO_2. Brown, Raman, Schlader, and Stannard (2013) definen la eficiencia ventilatoria como la relación lineal existente entre la producción de CO_2 y la respuesta ventilatoria.

Desde nuestro punto de vista, a estas definiciones se les puede incluir un matiz que complemente y de más información sobre el concepto de eficiencia ventilatoria. En nuestro caso definiríamos eficiencia ventilatoria como:

"Relación lineal existente entre la ventilación y la producción de CO_2, caracterizada por la magnitud del incremento en la respuesta ventilatoria de un sujeto por cada incremento que se produce en la producción de CO_2"

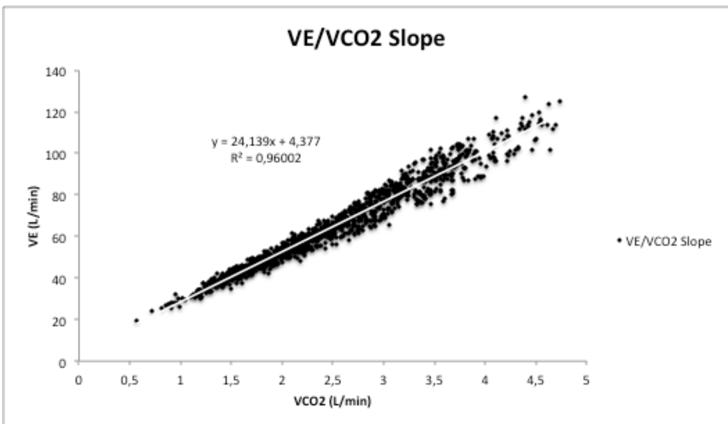


Fig 4: Relación entre la ventilación (VE) y la producción de CO_2 (VCO_2) en ciclistas de clase mundial.

De acuerdo con la definición propuesta, los sujetos más eficiente serían aquellos que obtuviera un menor incremento en su ventilación para un mismo incremento en la producción de CO_2.

2.1.2. Vías de evaluación de la eficiencia ventilatoria

1- VE/VCO$_2$ Slope o Delta CO$_2$

Esta medida de la eficiencia ventilatoria puede ser considerada como el "*Gold Standar*" ya que es usada en una gran cantidad de publicaciones científicas como vía para medir la eficiencia ventilatoria (Arena, Myers, et al., 2007; Brown et al., 2013; Schneider & Berwick, 1997; Sun et al., 2002; Ukkonen et al., 2008).

Si registramos el intercambio de gases durante un ejercicio de tipo incremental hasta el agotamiento, y representamos en un gráfico la ventilación en función de la producción de CO_2 mediante una dispersión de puntos, obtendremos una relación de tipo lineal entre ambas variables. El valor de la pendiente de la recta de regresión lineal ($y = a \cdot x + b$) es considerado como el valor de eficiencia ventilatoria de un sujeto (Fig. 4).

A nivel fisiológico, esta variable aporta información útil sobre la funcionalidad del sistema respiratorio y de una forma más concreta sobre su capacidad para eliminar CO_2 y normalizar el pH (Brown et al., 2013).

2- Oxygen Uptake Efficiency Slope (OUES)

En 1996, Baba et al. (1996) introdujeron el concepto de oxygen uptake efficiency slope (OUES). La OUES representa la magnitud del incremento del consumo de O_2 en respuesta a una ventilación dada durante un ejercicio de tipo incremental. Este parámetro aporta información acerca de cómo de efectiva es la captación 0_2 y la introducción del mismo en el organismo (Baba et al., 1996).

OUES se calcula mediante la representación del consumo de O_2 y el logaritmo de la ventilación medidos durante un ejercicio incremental, o lo que es lo mismo $VO_2=\log_{10} VE + b$. El valor de la pendiente de la recta de regresión lineal se corresponde con el valor de OUES (Fig. 5) (Akkerman et al., 2010).

Aunque se ha sugerido el uso de la OUES como indicador de eficiencia ventilatoria (Akkerman et al., 2010; Baba et al., 1996; Brown et al., 2013), desde nuestro punto de vista no es el método más adecuado. Las variaciones en la concentración de O_2 no tienen efecto directo alguno sobre el centro respiratorio central, en lo que se refiere a modificar el impulso respiratorio. Gracias al efecto amortiguador de la hemoglobina, el suministro de O_2 a los tejidos es el mismo, incluso cuando la PO_2 varía. Por lo tanto, excepto en condiciones especiales, el aporte de oxígeno es siempre adecuado, pese a las variaciones producidas en la ventilación (Hall & Guyton, 2001).

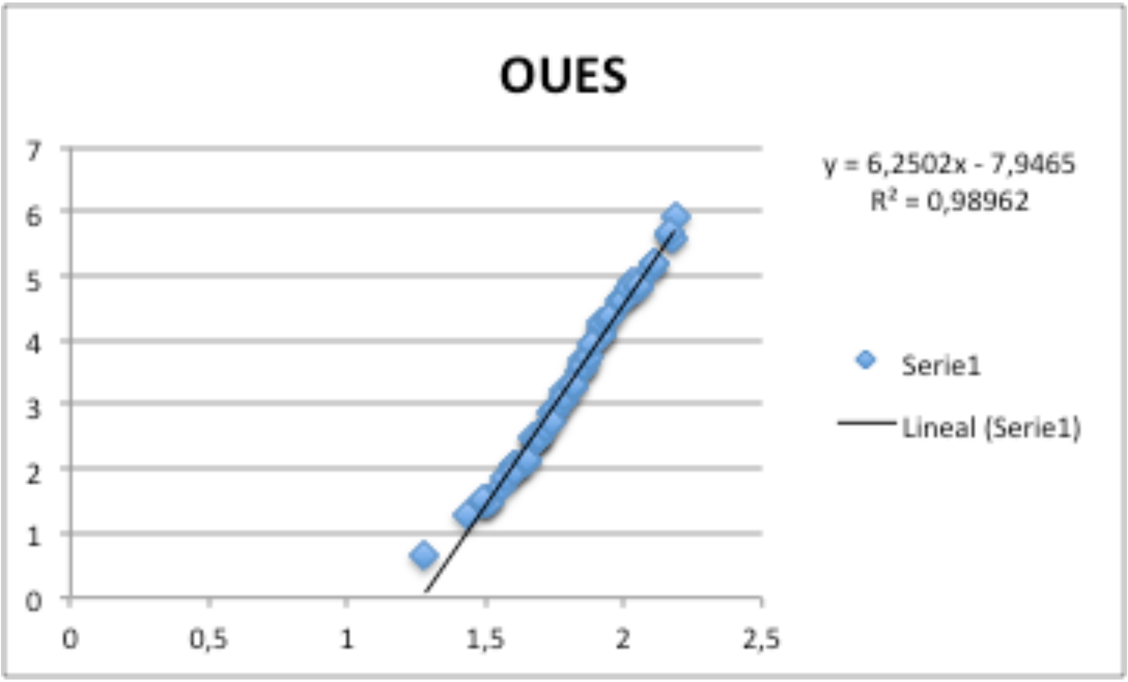


Fig. 5: Representación gráfica de la OUES en un ciclista de clase mundial

3- Equivalente Ventilatorio del CO_2

Otra propuesta para la evaluación de la eficiencia ventilatoria es la relacionada con el equivalente ventilatorio de CO_2 (VE/VCO_2). De acuerdo con Sun et al. (2002) podemos tomar el valor más bajo registrado durante un test incremental hasta el agotamiento como un indicador de eficiencia ventilatoria. De esto modo, el sujeto más eficiente sería aquel que consiguiera un valor más bajo de VE/VCO_2 durante la prueba.

Davis et al. (2006) compararon la reproductibilidad de dos medidas de eficiencia ventilatoria (VE/VCO_2 slope y el valor más bajo de VE/VCO_2), no encontrando diferencias significativas entre ambas y obteniendo unos aceptables valores de coeficiente de correlación. Pero desde nuestro conocimiento este parámetro presenta una limitación importante. Solo aporta información útil sobre la eficiencia ventilatoria a una intensidad concreta (carga constante) o en un momento concreto de la prueba. Si queremos conocer la evolución de la relación entre la VE y el VCO_2 durante la prueba, este parámetro no nos aporta información suficiente.

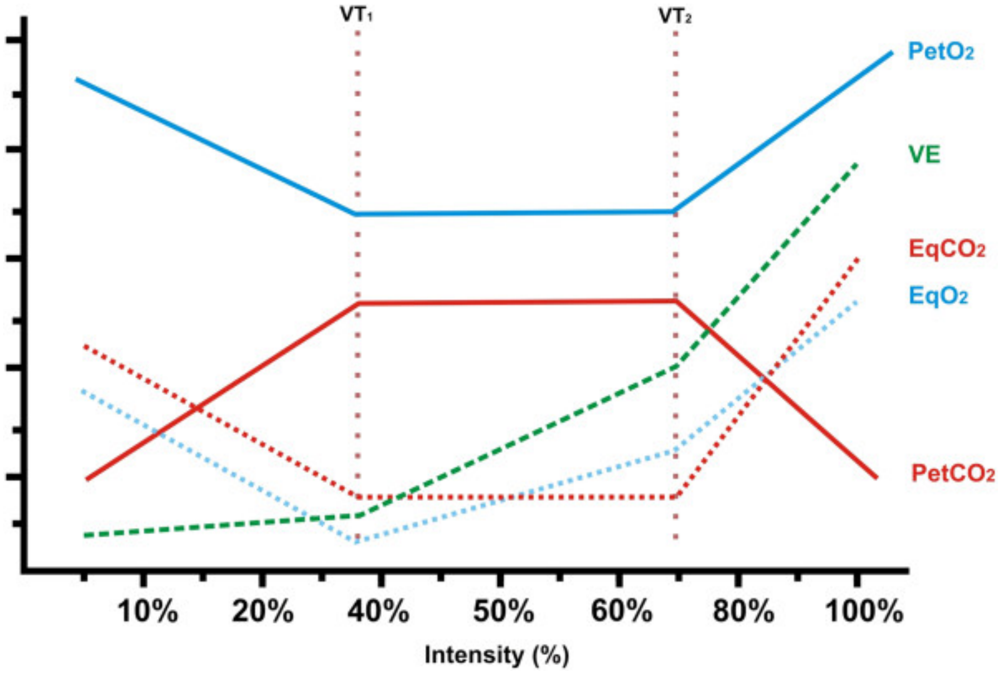


Fig. 6: Evolución del equivalente ventilatorio de O_2 (VE/VO_2) y el equivalente ventilatorio de CO_2 (VE/VCO_2) durante un ejercicio incremental (Skinner and Mclellan et al. 1980)

2.2. EFICIENCIA VENTILATORIA Y RENDIMIENTO DEPORTIVO

A continuación se expone una revisión bibliográfica de las publicaciones científicas relacionadas con la eficiencia ventilatoria y su aplicación al ámbito del rendimiento deportivo.

2.2.1. Eficiencia Ventilatoria en Sujetos Sanos

Si centramos nuestro objetivo en conocer cuál es el comportamiento de la eficiencia ventilatoria en poblaciones que no sufren ningún tipo de patología, encontramos que la respuesta de la misma tiende a permanecer constante. La respuesta de la eficiencia ventilatoria parece ser independiente del sexo (Guerrero, Naranjo, & Carranza, 2008), edad (Sun et al., 2002) o posición corporal en la que ésta sea evaluada (Terkelsen, Clark, & Hillis, 1999).

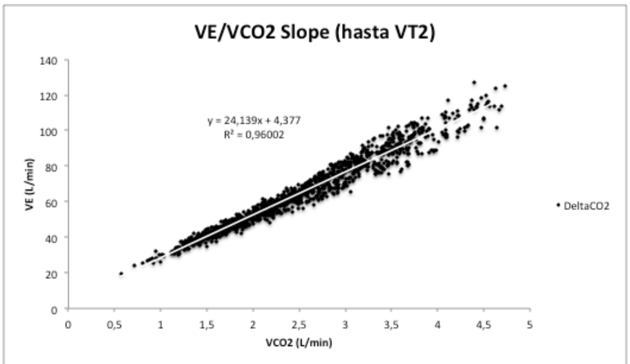
De acuerdo con Sun et al. (2002) los valores de la VE/VCO_2 slope se mueven en un rango de entre 22 y 27 en sujetos de <20 años hasta >60 años. Habedank et al. (1998) concluyeron que, como el consumo de O_2 (VO_2), la eficiencia ventilatoria es diferente en hombre y mujeres, tendiendo a reducirse con la edad en sujetos sanos. Valores superiores a 34 son indicativos de un mala eficiencia del sistema respiratorio, pudiendo llegar a considerarse como patológicos (Brown et al., 2013).

Entre los motivos que podrían justificar esta gran variabilidad intra-sujetos de la VE/VCO_2 slope se encuentra la aparente estabilidad del sistema de control de la respiración encargado de promover patrones respiratorios eficientes. Existen evidencia científica que sostiene que el entrenamiento no es capaz de modificar el patrón respiratorio de deportistas entrenados (Lucia, Carvajal, Calderon, Alfonso, & Chicharro, 1999; Lucia, Hoyos, Pardo, & Chicharro, 2001)

De acuerdo con lo anterior podemos suponer que cada sujeto posee un valor característico y personal de VE/VCO_2 slope, el cual no es modificable y tienda a comportarse de manera estable. Aunque este hecho ha de ser justificado por la literatura científica.

En segundo motivo que podría justificar esta gran variabilidad intra-sujetos es el punto hasta el cual la VE/VCO_2 slope ha sido evaluada. Si evaluamos la eficiencia ventilatoria incluyendo datos hasta el final del test incremental-agotamiento (por encima de VT_2) debemos de tener en cuenta que a partir de este punto la respuesta de la ventilación sufre un aumento considerable, lo que originará valores de VE/VCO_2 slope más elevados que si solo tomamos valores de VE Y VCO_2 hasta VT_2 (Fig. 7-8)

Pero desde nuestro conocimiento, no existen artículos científicos que evalúen la eficiencia ventilatoria en deportistas y que realicen un descripción de la misma en este tipo de población.



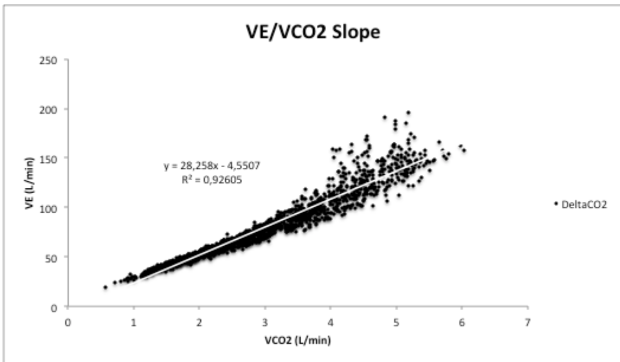


Fig. 7-8: Comparativa en la respuesta de la eficiencia ventilatoria medida como VE/VCO_2 Slope hasta el segundo umbral ventilatorio (VT_2) y hasta el agotamiento.

2.2.2. Eficiencia Ventilatoria y Entrenamiento

Pero si focalizamos nuestra atención en artículos científicos que evalúen la influencia de algún programa de entrenamiento sobre la eficiencia ventilatoria, encontramos que éstos solo han sido aplicados en poblaciones patológicas. Principalmente son sujetos con deficiencias cardiorrespiratorias los que consiguen una reducción en los valores de VE/VCO_2 slope gracias a la influencia de diferentes programas de entrenamiento.

Tras un periodo de entrenamiento específico de los músculos inspiratorios (IMT), el cual fue añadido a un programa de entrenamiento aeróbico (AT), se consiguió una mejora de la respuesta cardiorespiratoria en pacientes con fallo cardiaco (CHF) y debilidad inspiratoria (IMW). Se reportaron mayores mejoras en el grupo que incluyó IMT. El valor de la VE/VCO_2 Slope se redujo de 44±5 a 30±7 (Akkerman et al., 2010).

De igual modo, sujetos que presentaban CHF y valores iniciales de VE/VCO_2 slope de 31 (23-35,1) consiguieron reducir significativamente estos valores tras un periodo de IMT (Palau et al., 2014)

Con relación a el entrenamiento interválico de alta intensidad (HIIT), éste se mostró más efectivo que el entrenamiento continuo (MIT) sobre la mejora de la eficiencia ventilatoria de pacientes con CHF (Cardozo, Oliveira, & Farinatti, 2015). En este misma línea, se demostraron mejoras en marcadores de eficiencia ventilatoria tras un periodo de entrenamiento HIIT en pacientes con CHF, aunque no se reportaron cambios en la VE/VCO_2 Slope.

Por otro lado, Zavin et al. (2013) evaluaron la relación existente entre la fuerza muscular y la eficiencia ventilatoria, exponiendo que aquellos sujetos con menor fuerza muscular presentaban una menor eficiencia ventilatoria en la recuperación pos-ejercicio. Gonzales, Tucker, Kalasky, and Proctor (2012) encontraron correlaciones entre la fuerza muscular y la eficiencia ventilatoria en mujeres ancianas, reportando que aquellas mujeres que presentaban una menor fuerza de los mientras inferiores también presentaban mayores valores de ineficiencia ventilatoria.

Desde nuestro conocimiento no existen publicaciones científicas que traten de evaluar la posible variabilidad de la eficiencia ventilatoria en deportistas tras un periodo de entrenamiento controlado de los músculos inspiratorios o de un protocolo de entrenamiento interválico de alta intensidad.

2.2.3. Eficiencia ventilatoria y eficiencia muscular (DE, GE)

Se define a la *Gross Efficiency* (GE) o eficiencia mecánica como la relación (el cociente) entre el trabajo alcanzado·min^{-1} (vatios convertidos a kcal·min^{-1}) y la energía gastada·min^{-1} (en kcal·min^{-1}) para realizarlo (Coyle, Sidossis, Horowitz, & Beltz, 1992). Esta eficiencia mecánica no representa exactamente la eficiencia del la contracción muscular de los músculos involucrados en el pedaleo debido a que, al estar calculada a partir del VO_2 total en ejercicio, en ella también están contenidos el coste energético de otros procesos metabólicos que no contribuyen a la realización del trabajo, como por ejemplo el ritmo metabólico basal (Coyle et al., 1992; Gaesser & Brooks, 1975), el coste del trabajo de los músculos estabilizadores y de la musculatura respiratoria (Ettema & Lorås, 2009) y el coste del movimiento de los miembros inferiores (Ettema & Lorås, 2009; McDaniel, Durstine, Hand, & Martin, 2002; Sidossis, Horowitz, & Coyle, 1992). Por este motivo, se ha sugerido que la GE sí es una buena manera de medir eficiencia del todo el cuerpo (Coast & Welch, 1985). Esto, unido a la escasa variabilidad, la GE muestra (en comparación con la DE), hace que la GE haya sido muy estudiada en los últimos años (Coyle et al., 1992; Lucia, Hoyos, Pérez, Santalla, & Chicharro, 2002; Santalla, Naranjo, & Terrados, 2009).

La *Delta Efficiency* (DE) o delta eficiencia se ha definido como el cambio de la relación entre el trabajo realizado y el gasto energético, y se calcula como la pendiente de la regresión lineal (y=a·x+b) de la relación entre el gasto energético·min^{-1} (y, en kcal·min^{-1}), y el trabajo alcanzado·min^{-1} (x, en kcal·min^{-1}) (Coyle et al., 1992). La DE es una mejor estimación de la eficiencia a nivel muscular que la Gross Efficiency porque, el cambio en la energía gastada·min^{-1} es calculado sólo en base al cambio en el trabajo realizado·min^{-1}, por lo que (al contrario de la GE) elimina la influencia de los procesos metabólicos que no contribuyen a la realización del trabajo anteriormente mencionados (Coyle et al., 1992; Gaesser & Brooks, 1975; Santalla et al., 2009; Stainbsy, Gladden, Barclay, & Wilson, 1980). A pesar de que la DE suponga el aislamiento del coste energético de la musculatura implicada en el pedaleo, no puede considerarse como una valoración exacta sino como una estimación, ya que la variación de VO_2 en relación a la variación de carga no se debe solamente a los procesos relacionados directamente con la producción de fuerza (estos son los puentes cruzados de miosina). Otros procesos no relacionados con los puentes cruzados y dependientes del ATP, llamados "actividades de calor" (liberación y recaptación de calcio del retículo sarcoplásmico) podrían incluso afectar a la relación entre el VO_2 y el trabajo realizado (Barstow, Jones, Nguyen, & Casaburi, 2000).

De hecho, la gran variabilidad de la DE en pedaleo (10-25 %) (Ettema & Lorås, 2009) nos indica que el resto de la energía obtenida del la hidrólisis del ATP se usa para la homeostasis y para el calor. A pesar de todo esto, la DE sí puede dar una estimación razonable de la eficiencia muscular y ha sido propuesta como una forma válida para estimarla (Coyle et al., 1992; Gaesser & Brooks, 1975).

Los estudios más recientes realizados en ciclistas profesionales de clase mundial, tanto transversales (Lucia et al., 2002), como longitudinales (Coyle, 2005; Santalla et al., 2009) han arrojado evidencia de que el entrenamiento incrementa la eficiencia. Así, Lucia et al. (2002) describieron en un estudio transversal una correlación inversa entre el VO_2max y la GE en ciclistas profesionales de clase mundial (participantes habituales en el Tour de Francia, con varias victorias parciales obtenidas algunos de los sujetos), estableciendo la GE como un posible requisito para alcanzar/mantenerse en la alta competición y como un predictor de rendimiento en ciclismo profesional (los sujetos con mayor GE eran aquellos con mejores resultados en etapas contrarreloj (CRI) y en las clasificaciones generales de Giro, Tour y Vuelta).

De forma longitudinal, Coyle (2005) describió por primera vez el incremento de DE durante un periodo de 7 años en ciclismo profesional. El hecho de que se trate de un case report (con el 7 veces ganador del Tour de Francia Lance Armstrong), unido a las mas que duras críticas metodológicas recibidas y la relación de Lance en asuntos de dopaje, impiden extrapolar sus conclusiones.

Sin embargo, resultados previos describieron un incremento de DE en ciclistas profesionales de clase mundial (esto es: Ganadores de etapas, vueltas por etapas de 3 semanas y campeonatos del mundo) durante 5 años (Santalla et al., 2009). Además se encontró una correlación inversa DE-VO_2max (al final de los 5 años) similar a la encontrada por Lucia et al. (2002) entre la GE-VO_2max. Esto, evidencia no sólo un incremento de la eficiencia por entrenamiento acumulado, sino también que este incremento es una estrategia de selección/adaptación de los ciclistas dentro de la máxima categoría mundial. En ambos estudios, la posible modificación de la actividad de la ATPasa y del porcentaje de fibras tipo I por los años de entrenamiento acumulados son los argumentos sugeridos como responsables del incremento de eficiencia. Por su carácter invasivo, la ausencia biopsias musculares en estudios con ciclistas de clase mundial limita la demostración de este argumento. Sin embargo el mayor % de fibras tipo I densidad capilar/mitocondrial en ciclistas con 7 años de experiencia profesional (vs ciclistas con 3 años) (Rodríguez et al., 2002) apunta cómo posible dicho argumento. Resultados similares han sido obtenidos transversalmente (Hopker, Coleman, & Wiles, 2007) .

Pero desde nuestro conocimiento, no existen estudios científicos que evalúen cambios en la eficiencia ventilatoria a lo largo del tiempo.

Tampoco existen estudios que centren su atención en evaluar si cambios en el rendimiento deportivo pudieran estar relacionados con cambios en la eficiencia ventilatoria de deportistas entrenados.

2.2.4. Eficiencia ventilatoria e hipoxia

Si focalizamos nuestra atención en conocer que ocurre con la eficiencia ventilatoria en condiciones ambientales donde el aporte de 0_2 es limitado (condiciones de hipoxia), encontramos que no existen muchos estudios que hayan relacionado estas dos variables.

Bernardi, Schneider, Pomidori, Paolucci, and Cogo (2006) describieron las características fisiológicas de alpinistas de élite, comparando a aquellos que conseguían alcanzar la cima con éxito y aquellos que no. Estos autores reportaron diferencias entre ambos grupos, concluyendo que los alpinistas que conseguían cima eran aquellos que presentaban una mayor eficiencia ventilatoria y menor respuesta ventilatoria en hipoxia (HVR).

Esposito, Limonta, Alberti, Veicsteinas, and Ferretti (2010) compararon la influencia de un programa de entrenamiento IMT, sobre la potencia aeróbica en condiciones de normoxia e hipoxia. Se encontraron mejoras en la función respiratoria, aunque estas mejoras no consiguieron provocar cambios en el VO_2max en ninguna de las dos situaciones anteriormente mencionadas.

Pero desde nuestro conocimiento, no existen publicaciones científicas que hayan evaluado posibles cambios en la eficiencia ventilatoria en condiciones de hipoxia.

En esta misma línea, tampoco tenemos constancia de la existencia de publicaciones científicas que evalúen la influencia de un programa de entrenamiento de los músculos inspiratorios sobre la eficiencia ventilatoria en condiciones de hipoxia.

CAPÍTULO 3

HIPÓTESIS / HYPOTHESIS

SALAZAR-MARTÍNEZ E.

3- HIPÓTESIS

La VE/VCO_2 slope ha sido usada tanto en el ámbito clínico (Arena, Guazzi, et al., 2007; Arena, Myers, et al., 2007; Ingle et al., 2007), como en sujetos sanos (Sun et al., 2002). De hecho, se sabe que los valores de VE/VCO_2 slope varían entre 19 y 32 en sujetos sanos (Sun et al., 2002), siendo aquellos valores superiores a 34 considerados como anormales (Arena, Guazzi, et al., 2007; Arena, Myers, et al., 2007) o indicativos de ineficiencia del sistema respiratorio (Brown et al., 2013). En este sentido, en atletas entrenados la VE/VCO_2 slope podría aportar información útil acerca de su capacidad para eliminar CO_2 y regular el pH sanguíneo durante el ejercicio. Se podría suponer que un gran volumen de entrenamiento/competición con alta exigencia cardiorrespiratoria debería desarrollar una buena VE/VCO_2 slope del mismo modo que lo hace con otros parámetros (Lucia, Pardo, Durantez, Hoyos, & Chicharro, 1998; Sallet, Mathieu, Fenech, & Baverel, 2006).

A diferencia de la eficiencia ventilatoria, el patrón respiratorio sí ha sido ampliamente estudiado en deportistas (Benchetrit, 2000; Lucia et al., 1999; Lucia et al., 2001; Scheuermann & Kowalchuk, 1999). Una forma sencilla de analizar el patrón respiratorio es evaluar las relaciones entre el volumen tidal (VT) y la frecuencia respiratoria (BF) (Milic-Emili, G. & Cajani, 1957). Pero desde los años 70 (Milic-Emili, J., 1982; Milic-Emili, J. & Grunstein, 1976), sabemos que la VE puede ser descompuesta en el producto de dos componentes que aportan mucha más información: a) la actividad inspiratoria central, conocida como "driving" y expresada por la relación entre el VT y el tiempo inspiratorio (VT/Ti) y b) el mecanismo de alternancia inspiración-espiración, conocido como "timing" y expresado por la relación entre el Ti y la duración total del ciclo respiratorio (Ti/Ttot). Analizar todas estas variables (VE, VT, BF, VT/Ti, Ti/Ttot) ha sido la vía más usada para evaluar el patrón respiratorio tanto en sujetos sedentarios (Benchetrit, 2000) como en atletas (Lucia et al., 2001; Lucia, Hoyos, Santalla, Earnest, & Chicharro, 2003). Se podría también pensar que un mayor desarrollo de la VE/VCO_2 slope podría estar relacionado con un buen control de la respiración y con un patrón respiratorio efectivo.

En atletas entrenados se sabe que la musculatura inspiratoria, como cualquier músculo estriado, experimenta cambios y adaptaciones con el entrenamiento, mejorando su fuerza y reduciendo su fatiga (Dempsey et al., 1996; Romer, McConnell, & Jones, 2002; Tong et al., 2008). En músculo esquelético estos cambios se reflejan en la mejora de la eficiencia muscular, medida a través de la *Delta Eficiencia* (DE) (Coyle et al., 1992). Así, se han descrito cambios longitudinales en la DE debidos a las adaptaciones provocadas por el entrenamiento y la competición en ciclistas profesionales de clase mundial a lo largo de varias temporadas competitivas (Santalla et al., 2009). Del mismo modo, se han descrito transversalmente que la eficiencia mecánica global o *Gross Efficiency* (GE) es mayor en ciclistas profesionales comparada con ciclistas aficionados (Sallet et al., 2006). Incluso se ha sugerido un aumento de los valores GE y DE en los ciclistas profesionales de clase mundial como estrategia de rendimiento en alta competición (Lucia et al., 2002; Santalla et al., 2009).

Dado que la eficiencia muscular y mecánica cambia a lo largo del tiempo en deportistas altamente entrenados en resistencia, sería lógico pensar que también pudiera hacerlo la eficiencia respiratoria, medida como VE/VCO_2 slope. Sin embargo, entre los pocos trabajos que estudian esta variable en ejercicio, existen indicios para suponer que el valor de la VE/VCO_2 slope tiende a permanecer constante (Sun et al., 2002). En ejercicio no se han encontrado diferencias significativas en la eficiencia ventilatoria entre sexos (Guerrero et al., 2008), grupos de edad (Sun et al., 2002) o la posición corporal (Terkelsen et al., 1999). Tampoco se han descrito cambios en la eficiencia ventilatoria tras un periodo de entrenamiento no controlado de 16 semanas en ciclistas juveniles (Brown et al., 2013). Por el contrario, en poblaciones patológicas si se han descrito cambios tras la aplicación de programas de entrenamiento (Cardozo et al., 2015; Gonzales et al., 2012; Myers et al., 2012; Palau et al., 2014). Sin embargo, todos estos estudios han sido realizados en sujetos con patologías y moderadamente entrenados y no en deportistas. Podría ser posible que un protocolo de entrenamiento de los músculos respiratorios mejore la respuesta de la eficiencia ventilatoria en normoxia e hipoxia gracias a una reducción de las demandas metabólicas de estos músculos. En esta misma línea, un entrenamiento interválico de alta intensidad podría provocar cambios en la respuesta de la eficiencia ventilatoria de deportistas gracias a ajustes en el patrón respiratorio y los grandes demandas ventilatorias desarrolladas durante este tipo de protocolo de entrenamiento.

De acuerdo con todo lo anterior, las hipótesis planteadas para la presente Tesis Doctoral Internacional son:

1. La eficiencia ventilatoria podría ser un factor determinante del rendimiento deportivo en sujetos sanos.

2. Cambios en el rendimiento deportivo podrían estar relacionados con cambios o mejoras en la eficiencia ventilatoria.

3. Cambios en la eficiencia ventilatoria podrían estar relacionados con cambios en el patrón respiratorio.

4. La eficiencia ventilatoria podría comportarse de manera similar en deportistas independientemente de las características de cada deportista.

5. La eficiencia ventilatoria podría variar en condiciones de normoxia e hipoxia.

6. Un protocolo de entrenamiento de la musculatura inspiratoria podría promover o mejorar la respuesta de la eficiencia ventilatoria en deportistas.

7. El entrenamiento interválico de alta intensidad podría provocar cambios en la eficiencia ventilatoria en deportistas.

CAPÍTULO 4

OBJETIVOS / AIMS

SALAZAR-MARTÍNEZ E.

4- OBJETIVOS

A continuación se especifican los objetivos que fueron establecidos para el desarrollo de esta Tesis Doctoral Internacional y los estudios científicos se encuentran materializados:

A. Conocer la relación entre la eficiencia ventilatoria y el rendimiento deportivo en deportistas (**estudios I, II, III, IV**).

B. Comprender en mejor medida la influencia del centro respiratorio en la respuesta de la eficiencia ventilatoria durante el ejercicio (**estudios I, IV**).

C. Conocer si existen diferencias en la respuesta de la eficiencia ventilatoria en deportistas de diferentes disciplinas, características antropométricas, nivel de condición física y edad (**estudio IV**).

D. Aportar una mayor evidencia científica sobre la respuesta de la eficiencia ventilatoria en ejercicio incremental en deportistas (**estudios I, II, III, IV**).

E. Conocer el comportamiento de le eficiencia ventilatoria en condiciones de hipoxia (FiO_2=16.45) y normoxia (FiO_2=21) (**estudio II**).

F. Evaluar la influencia del entrenamiento de la musculatura respiratoria sobre la eficiencia ventilatoria de sujetos sanos (**estudio II**).

G. Demostrar si el entrenamiento de la musculatura inspiratoria mejora la respuesta de la eficiencia ventilatoria en condiciones de hipoxia (**estudio II**).

H. Conocer si el entrenamiento de la musculatura inspiratoria mejora el rendimiento en condiciones de hipoxia (**estudio II**).

I. Evidenciar si mejoras en el rendimiento deportivo tras un periodo de entrenamiento de la musculatura respiratoria están relacionadas con mejoras en la eficiencia ventilatoria en deportistas (**estudio II**).

J. Comparar el comportamiento de la eficiencia ventilatoria en condiciones de hipoxia y normoxia (**estudio II**).

K. Conocer la respuesta de la eficiencia ventilatoria tras la aplicación de un programa de entrenamiento (**estudio III**).

L. Evidenciar si los posibles cambios en el rendimiento deportivo tras la aplicación de un programa de entrenamiento interválico de alta intensidad de 3 semanas están asociados o relacionados con una modificación de la eficiencia ventilatoria (**estudio III**).

CAPÍTULO 5

METODOLOGÍA / METHODOLOGY

SALAZAR-MARTÍNEZ E.

5- METODOLOGÍA

5.1.- PLAN DE ACCIÓN GENÉRICO

Para la consecución de los objetivos anteriormente mencionados se emplearon las siguientes técnicas metodológicas de investigación:

1- Estudio Observacional-Descriptivo

Gracias a la relación de colaboración establecida hace algunos años entre nuestro grupo de investigación y un equipo ciclistas UCI-PRO Tour, disponíamos en nuestra base de datos registros de pruebas de esfuerzos realizadas a ciclistas profesionales de clase mundial durante varios años consecutivos.

Normalmente es difícil estudiar ciclistas del más alto nivel debido a la cantidad de horas de entrenamiento y competición que acumulan a lo largo de las temporadas competitivas, pero gracias a las evaluaciones que nuestro grupo de investigación realizó durante varias temporadas a un equipo UCI-Pro Tour, a los que se había calculado la DE (Santalla et al., 2009), pudimos analizar de forma retrospectiva parámetros respiratorios de estos ciclistas de clase mundial.

Nuestro objetivo fue indagar en estos datos, dada su gran potencialidad, y llevar a cabo un estudio descriptivo sobre la respuesta de la eficiencia ventilatoria en estos atletas a lo largo del tiempo (estudio I).

2- Estudio Experimental

Para esclarecer las hipótesis anteriormente planteadas, se llevaron a cabo dos protocolos experimentales. En uno de ellos se introdujo un protocolo de entrenamiento de la musculatura inspiratoria (estudio II). En el otro se llevó a cabo un protocolo interválico de alta intensidad de 3 semanas (estudio III) a estudiantes de ciencias de la actividad física y deporte.

En el estudio II, se distribuyeron a los sujetos de forma aleatoria y randomizada en dos grupos diferentes (experimental y control). Al grupo experimental se le implemetó un protocolo de entrenamiento respiratorio consistente en 30 respiraciones usando un Power Breathe, con una carga del 50% de la presión inspiratoria máxima (50%P_{imax}), 2 veces al día, 5 días a la semana, durante un periodo de 6-8 semanas. El grupo control no llevó a cabo actividad física durante el periodo de estudio. Este tipo de entrenamiento es el más usado y recomendado por la bibliografía científica relacionada con la materia (Illi, Held, Frank, & Spengler, 2012; McConnell & Romer, 2004; Sheel, 2002).

Antes de la inclusión del protocolo experimental los sujetos completaron unos test iniciales de valoración. Los test consisitieron en la realización de una prueba de esfuerzo incremental con análisis de gases, un espirometría y un test de rendimiento a carga constante al 85% del VO_2max. Los test fueron realizados en condiciones de normoxia e hipoxia. Estos mismos test fueron repetidos una vez finalizado el proceso experimental.

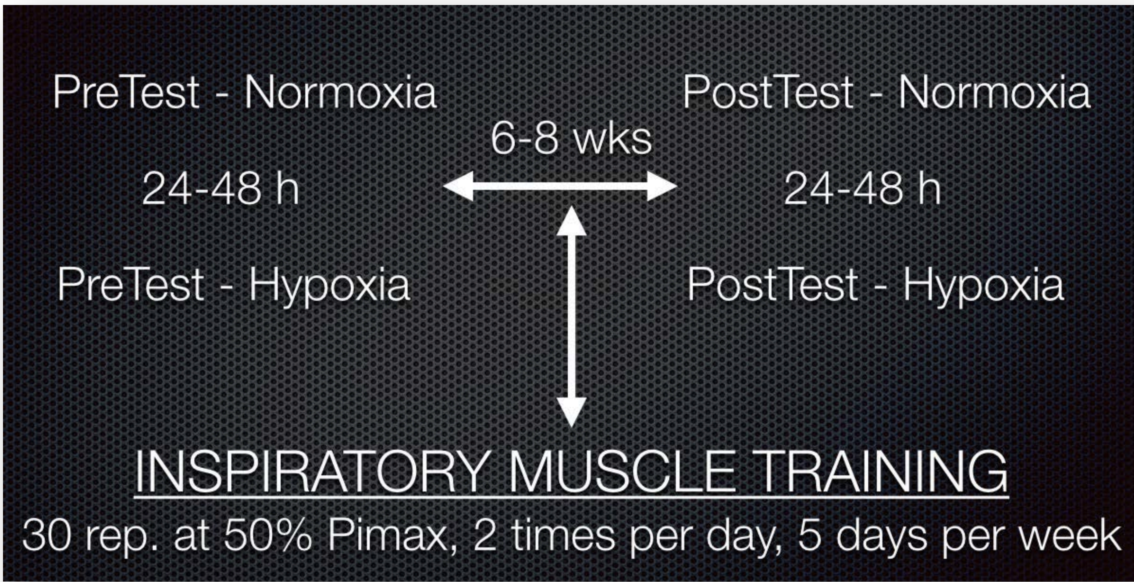


Fig. 9: Resumen del protocolo experimental desarrollado en el estudio II

En el estudio III, los sujetos completaron un test incremental con análisis de gases una vez incluidos en el estudio. A continuación, se distribuyeron a los sujetos en dos grupos (experimental y control) en función de su VO_2max. El grupo experimental completó 3 semanas de intervención compuesta de: 3 sesiones semanales de series de 4 min al 90-95% VO_2max. El grupo control mantuvo su nivel habitual de entrenamiento, sin la inclusión de nuevas tareas.

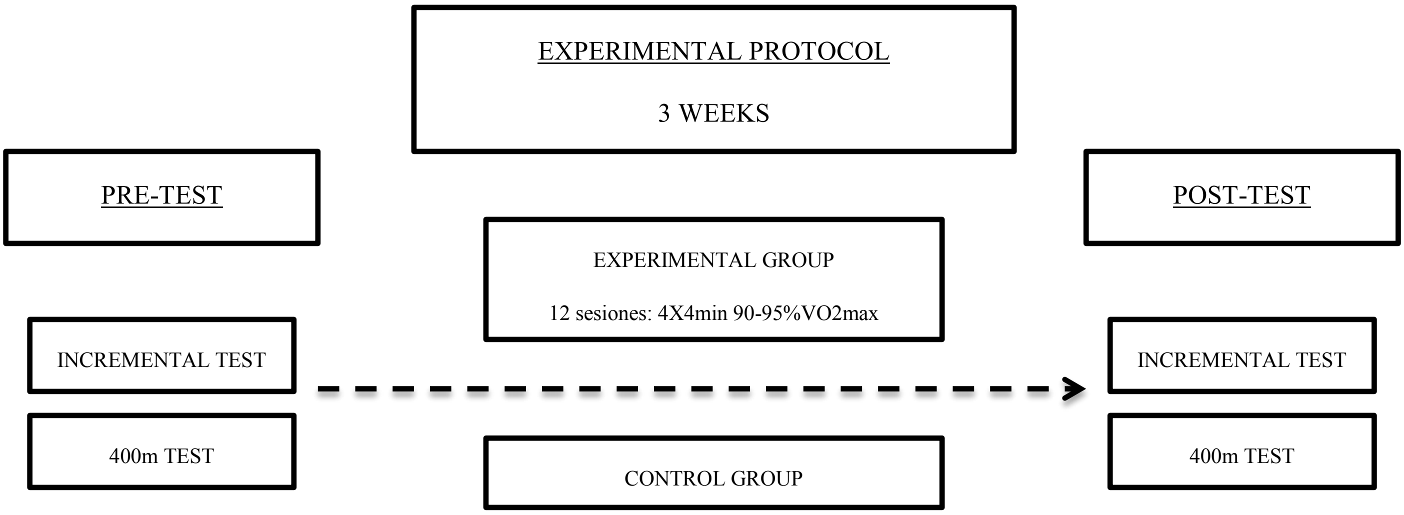


Fig. 10: Resumen del protocolo experimental desarrollado en el estudio III

3.- Estudio Correlacional

Una vez llevados a cabo los test iniciales, el protocolo experimental y los test finales, se llevó a cabo una análisis correlacional entre variables. Este análisis tuvo como objeto conocer la relación existente entre la eficiencia ventilatoria y variables de rendimiento deportivo (estudios I, II, III, IV).

CAPÍTULO 6

RESULTADOS Y DISCUSIÓN / RESULTS AND DISCUSSION

SALAZAR-MARTÍNEZ E.

6- RESULTADOS Y DISCUSIÓN [Results and Discussion]

La sección de resultados y discusión ha sido desarrollada específica e individualizadamente en cada uno de los capítulos de los que esta Tesis Doctoral está compuesta. A continuación se exponen cada uno de ellos:

ESTUDIO I

VENTILATORY EFFICIENCY AND BREATHING PATTERN IN WORLD-CLASS CYCLISTS: A THREE-YEAR OBSERVATIONAL STUDY

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VENTILATORY EFFICIENCY AND BREATHING PATTERN IN WORLD-CLASS CYCLISTS: A THREE-YEAR OBSERVATIONAL STUDY

ABSTRACT

The purpose of this three-year observational study was to analyze the ventilatory efficiency and breathing pattern in world-class professional cyclists. Twelve athletes (22.61±3.8 years; 177.38±5.5 cm; 68.96±5.5 kg and VO_{2max} 75.51±3.3mL·kg^{-1}·min^{-1}) were analyzed retrospectively. For each subject, respiratory and performance variables were recorded during incremental spiroergometry: oxygen uptake (VO_2), carbon dioxide output (VCO_2), pulmonary ventilation (VE), tidal volume (Vt), breathing frequency (f_R), driving (Vt/Ti), timing (Ti/Ttot), peak power output (PPO) and maximum oxygen uptake (VO_{2max}). Ventilatory efficiency (VE/VCO_2 slope) was calculated from the beginning of exercise testing to the second ventilatory threshold (VT_2). The VE/VCO_2 slope was unaffected during the study period (24.63±3.07; 23.61±2:79; 24:89±2:61) with a low effect size (ES=0.04). The PPO improved significantly in the third year (365±33.74; 386.36±32.33; 415.00±24.15) ($p < 0.05$). The breathing pattern variables, Vt/Ti and Ti/Ttot, did not change significantly over the three year period (ES=0.00; ES=0.03 respectively). These findings suggest that changes in cycling performance in world-class professional cyclists do not modify breathing variables related to the control of ventilatory efficiency.

Keywords**:** ventilatory efficiency / breathing pattern / cyclists / ventilation / VE/VCO_2 slope

1. Introduction

Carbon dioxide (CO_2) is produced in cellular metabolism and expelled into the atmosphere by ventilation (VE), but during this process CO_2 plays a fundamental role in the regulation of bodily pH, vascular tone (Gilbert, 2005) and in the ventilation control (Milsom et al., 2004). The relationship between the rate of CO_2 output (VCO_2) and VE in different circumstances has been widely described as a measurement of breathing efficiency (Arena et al., 2007b; Arena et al., 2008; Sun et al., 2002) at a given metabolic rate. During incremental effort, the slope of the linear relationship between VE and VCO_2 (VE/VCO_2 slope or DeltaCO_2) is the most widely used method to evaluate ventilatory efficiency (Arena et al., 2007b; Brown et al., 2013; Schneider and Berwick, 1998; Sun et al., 2002; Ukkonen et al., 2008).

The VE/VCO_2 slope has been commonly used in patients suffering from congestive heart failure (Arena et al., 2007a; Arena et al., 2007b; Ingle et al., 2007; Laveneziana et al., 2010, Robertson, 2011) as well as in healthy subjects (Sun et al., 2002). It is well established that the values of the VE/VCO_2 slope vary from 19 to 32 in healthy subjects (Sun et al., 2002), with values exceeding 34 considered abnormal (Arena et al., 2007a; Arena et al., 2007b) or indicative of the inefficiency of the respiratory system (Brown et al., 2013). The large variability in VE/VCO_2 slope could be an inborn characteristic but it could also be explained by the lack of consensus of measurement methods. Thus, differences in the VE/VCO_2 slope arise depending on whether it is measured from rest to VT_2 or from rest to the maximal work load.

In trained athletes, the VE/VCO_2 slope has not been wide studied and its relationship with sport performance is unclear. It could be possible that two athletes had different values of equivalent of CO_2 (VE/VCO_2) to the same metabolic rate, but they show the same VE/VCO_2 slope throughout the entire incremental test. In this case, they have different efficiency to a given level but the same global efficiency. They need the same increase in VE for every increase of 1 $l\cdot min^{-1}$ in CO_2 production. It could be possible that the high demands of elite cycling promote a lower VE/VCO_2 slope, involving a lower increase in VE for a given increment in VCO_2. Conditions where the CO_2 production is elevated, such as cycling, seems to play an essential role in the ventilatory control (Milsom et al., 2004). The ventilatory efficiency control could change over time in presence of a large amount of training and competition as it happens with others respiratory and performance variables (Lucia et al., 1998; Sallet et al., 2006).

Accepted that the VE/VCO_2 slope in normal subjects is a marker of ventilatory sensitivity, there are three possible mechanisms by which respiratory efficiency could change with training. One is through changes in the dead space (VD) (Wood et al, 2008); the other is a better mechanical performance of respiratory muscles (Sheel, 2002) and the third is an improved sensitivity of chemoreceptors (Babb et al, 2010).

It may also be expected that changes in the VE/VCO_2 slope with training could be related to improvements of breathing control by a more effective breathing pattern. Unlike ventilatory efficiency, the breathing pattern has been widely studied in athletes (Benchetrit, 2000; Lucia et al., 1999; Lucia et al., 2001; Scheuermann and Kowalchuk, 1999). A simple way to analyze the breathing pattern is to evaluate the relationship between the tidal volume (Vt) and breathing frequency (f_R) (Milic-Emili and Cajani, 1957). However, since the 1970s (Milic-Emili, 1982; Milic-Emili and Grunstein, 1976), it has been known that VE can be decomposed into the product of two components which offer more information: a) central inspiratory activity, known as "driving" and expressed as the relationship between Vt and inspiratory time (Vt/Ti) and b) the inspiration-expiration alternation, known as "timing", and expressed by the relationship between Ti and the total duration of the breathing cycle (Ti/Ttot). The analysis of all these variables (VE, Vt, BF, Vt/Ti, Ti/Ttot) is nowadays the most widely-used method to evaluate the breathing pattern in patients (Beltrão et al., 2015), sedentary subjects (Benchetrit, 2000) and athletes as well (Lucia et al., 2001; Lucia et al., 2003).

The studies which evaluated the VE/VCO_2 slope in healthy people were performed with sedentary or moderately trained subjects but not in highly trained athletes. In addition, we are not aware of any articles dealing with longitudinal VE/VCO_2 slope observations in athletes. It is usually difficult to study cyclists of the highest level due to the number of hours of training and competition they undergo over the competitive seasons, but thanks to the evaluations carried out by our research group over several seasons with a UCI-Pro Tour team (Santalla et al., 2009), we were able to analyze all these respiratory variables of interest in world-class cyclists.

We hypothesized that the high demands of professional cycling could induce changes in ventilatory efficiency, measured as the VE/VCO_2 slope, in world-class cyclists over a three-year period. If true, we would expect that changes in the VE/VCO_2 slope are related to changes in breathing pattern.

Therefore, the purpose of this study was to perform a retrospective longitudinal evaluation of the ventilatory efficiency and breathing pattern in world-class cyclists.

2. Material and methods

2.1. Subjects

A total of 42 male world-class professional cyclists, tested during the same period of the season in the same laboratory for at least five seasons, were retrospectively analyzed to select those for whom consecutive evaluations were available over a period of at least three years. Finally, 12 cyclists (starting age 22.61 ± 3.8 years; height 177.38 ± 5.5 cm; body weight 68.96 ± 5.5 kg and 76.92 ± 5.9 $mL \cdot kg^{-1} \cdot min^{-1}$) were included who had participatedannually in at least one of the three-week stage races (Giro d'Italia, Tour de France and Vuelta a España) or were evaluated at least two times consecutively over three years at the same time point in the season. Some of the subjects were among the best cyclists in the world (one winner of the Tour de France, one winner of the Vuelta a España and first in the annual ICU world ranking, one three-time Tour de France Podium, two Vuelta a España Podium, one Junior World Time Trial Champion, and two one-week stage race winners). All subjects provided written informed consent before testing. The study has been approved by the ethics committee of the Pablo de Olavide University (Seville).

2.2. Exercise test

Tests were always conducted during the first phase of the cyclists' competitive season. All incremental exercise tests have been performed on the same electromagnetically braked cycle ergometer. This ergometer allowed the subjects to choose their own pedal frequency and to adopt a position similar to that on their bicycles (Orion S.T.E., Toulouse, France). The distances and dimensions for saddle, handlebars and connecting rod were monitored and remained constant during the entire test period. The test was started at a power output (PO) of 100 W, after which PO was increased by 50 W every 4 min until exhaustion. This exercise protocol has already been used in previous research (Fernandez-Garcia et al., 2000). The freely chosen pedaling cadence generally ranged from 77 to 115 revolutions per min (rpm). Heart rate was monitored using radio-telemetry (Sport tester PE 4000; Polar, Kempele, Finland). Ventilation and respiratory gases were measured continuously and the highest 30-s VO_2 value was considered as VO_{2max}. VO_2, VCO_2, VE, Vt, f_R, Ti, Te and Ttot were measured breath by breath (BxB) using a gas analyzer (Vmax 29; Sensormedics, Yorba Linda, CA), which was calibrated before every exercise session. During the data collection period (three years), neither the cycle ergometer nor the gas analyzer was replaced and all the equipment passed the maintenance procedures recommended by the manufacturers. The ergometer was calibrated by the manufacturers annually. In addition, all the tests were performed under similar ambient temperature conditions (20–24C and 45%–65% relative humidity).

2.3. Ventilatory efficiency and breathing pattern

The ventilatory efficiency of each subject was calculated from the slope of the relationship between VCO_2 and VE during each test. To exclude the influence due to respiratory compensation for acidosis during highly intensive exercise, the VE/VCO_2 slope was determined from the beginning of the test until the second ventilatory threshold (VT_2). The breathing pattern was evaluated by the analysis of Vt, f_R, Vt/Ti and Ti/Ttot. The value of the slope representing the relationship between VE and Vt/Ti during each test (the driving slope) was used to test the central component.

2.4. Statistical analysis

The data is expressed as mean ± SD for each variable. The normality of the data was checked by means of the Shapiro-Wilk test. The homogeneity of variance was evaluated by Levene´s test. To compare the values obtained for each variable during the three-year observation period, one-way ANOVA with repeated measurements and the Friedman F-test (nonparametric conditions) were used. When significant differences were found, the Bonferroni test was used as a post hoc test. Effect sizes (ES) were also calculated using Eta-Squared. Intra-class correlations (ICC) and Pearson correlation coefficient (Pearson-r) were used to determine the reproducibility of measurements over time for VE/VCO_2 slope, VO_{2max} and PPO.Correlation analyses were carried out between VO_{2max}, VE/VCO_2 slope and PPO. The level of significance was set at $p<0.05$ for each statistical analysis.

3. Results

Data on the ventilatory and performance variables evaluated are shown in Table 1. No significant differences were found in any of the respiratory variables studied. VE/VCO_2 slope and VO_{2max} show a weak effect size (ES=0.04; 0.03) respectively. Significant differences were found in the PPO between the first and the third year, whit a large effect size (ES=0.32). Table 2 shows the test-re-test reliability, with the ICC and Pearson-r for VE/VCO_2 slope, VO_{2max} and PPO measurements. Figure 1, 2 and 3 show the mean values obtained each year for VE/VCO_2 slope, VO_{2max} and PPO respectively. Figure 4 shows the correlation between VCO_2 and VE for each year. The overall value of VE/VCO_2 slopes obtained was similar to the means values found each year. Figure 5 and 6 demonstrates the relationship between VO_{2max}, VE/VCO_2 slope and PPO with the values of all subjects over the entire study period. No correlations were found between VE/VCO_2 slope and VO_{2max} and between VE/VCO_2 slope and PPO.

Table 1

Evolution of ventilatory and performance variables during the study period

	1^{st} year	2^{nd} year	3^{th} year	*p*-value	*Effect Size*
VE/VCO_2 slope	24.63±3.07	23.61±2.79	24.89±2.61	0.549	0.04
VT/Ti slope	30.79±1.59	31.09±1.29	30.86±1.38	0.883	0.00
Vt/Ti ($l \cdot seg^{-1}$) (up to PPO)	5.13±0.29	5.17±0.53	5.02±0.47	0.541	0.02
Ti/Ttot (up to PPO)	0.47±0.01	0.46±0.02	0.47±0.01	0.655	0.03
Vt (l) (up to PPO)	3.15±0.55	3.14±0.46	3.28±0.55	0.694	0.01
Vt (l) (up to VT2)	2.7±469	2.84±398	2.86±428	0.659	0.02
f_R (breaths$\cdot$min^{-1}) (up to PPO)	50.50±10.36	50.64±10.13	48.90±11.10	0.457	0.00
f_R (breaths$\cdot$min^{-1}) (up to VT2)	35±5.77	37±7.31	35.2±5.79	0.73	0.02
VO_{2max} ($mL \cdot kg \cdot min^{-1}$)	75.92±6.28	75.29±6.09	77.93±5.31	0.578	0.03
VE_{max} ($l \cdot min^{-1}$)	155.54±17.97	156.37±17.76	153.33±15.98	0.918	0.00
PPO (W)	365±33.74	386.36±32.33	415.00±24.15	0.011*	0.32*

VE/VCO_2 slope, ventilatory efficiency; Vt/Ti slope, driving; Ti/Ttot, timing; Vt, tidal volume; f_R breathing frequency; VO_{2max}, maximum oxygen uptake, VE, maximum ventilation, PPO, peak power output. All values are expressed as mean ± SD.

* Significantly different from 1^{st} year vs 3^{th} year in PPO ($p < 0.05$).

*Large effect size (ES ≥ 0.14)

* $p < 0.05$

Table 2

Test-re-test reliability between repeated measurements over three years study period

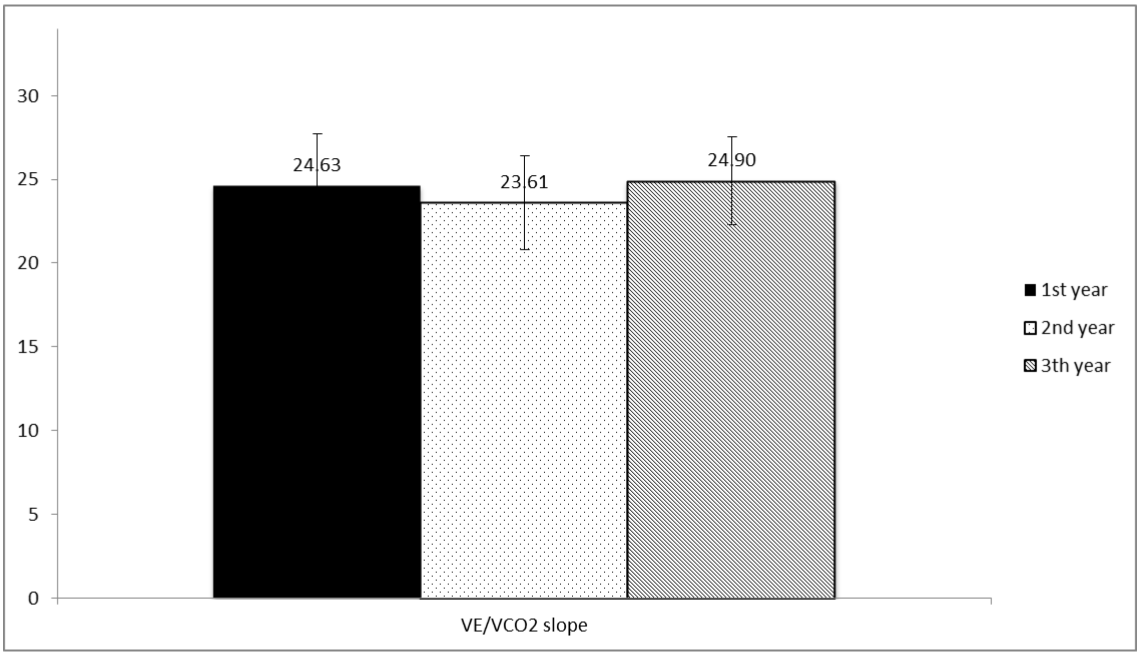
		Pearson r		
	CCI	*1st year - 2nd year*	*1st year - 3th year*	*2nd year-3th year*
VE/VCO_2 slope	0.896	0.900*	0.911*	0.745*
VO_{2max}($mL \cdot kg \cdot min^{-1}$)	0.234	0.543	-0.392	0.234
PPO (W)	0.029	0.688*	0.745*	0.378

CCI, correlation coefficient intra-class; VE/VCO_2 slope, ventilatory efficiency; VO_{2max}, maximum oxygen uptake, PPO, peak power output.

* Significantly correlation (p< 0.05).

4. Discussion

To the best of our knowledge, this is the first longitudinal study which analyzed the ventilatory efficiency (VE/VCO_2 slope) and breathing pattern in world-class cyclists. The main finding of the present study was that ventilatory efficiency, measured as VE/VCO_2 slope, and breathing pattern did not change in top cyclists over a three-year observation period. Our results agree with previous data suggesting that ventilatory efficiency would be a variable which is maintained within a constant range regardless of physical effort and training adaptations (Brown et al., 2013; Guerrero et al., 2008; Sun et al., 2002; Terkelsen et al., 1999).



Figigure 1: Means values of ventilatory efficiency (VE/VCO_2 slope) between years in world-class cyclists. No significant differences were found between years.

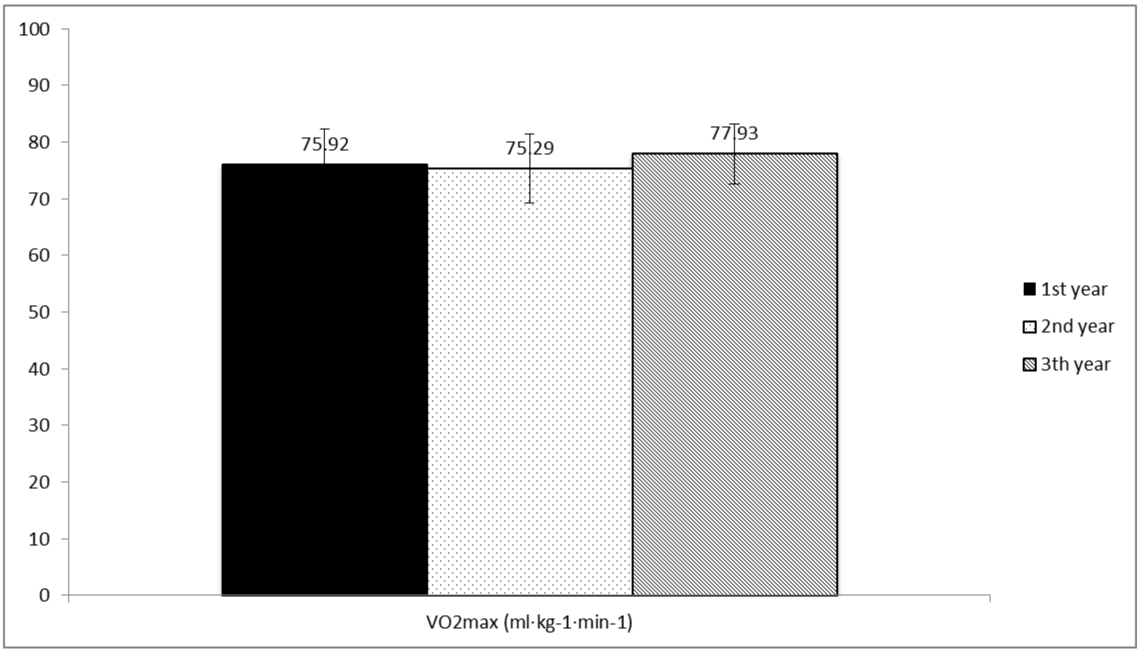


Figure 2: Means values of maximum oxygen uptake (VO_{2max}) over three years study period in world-class cyclists. No significant differences were found between years.

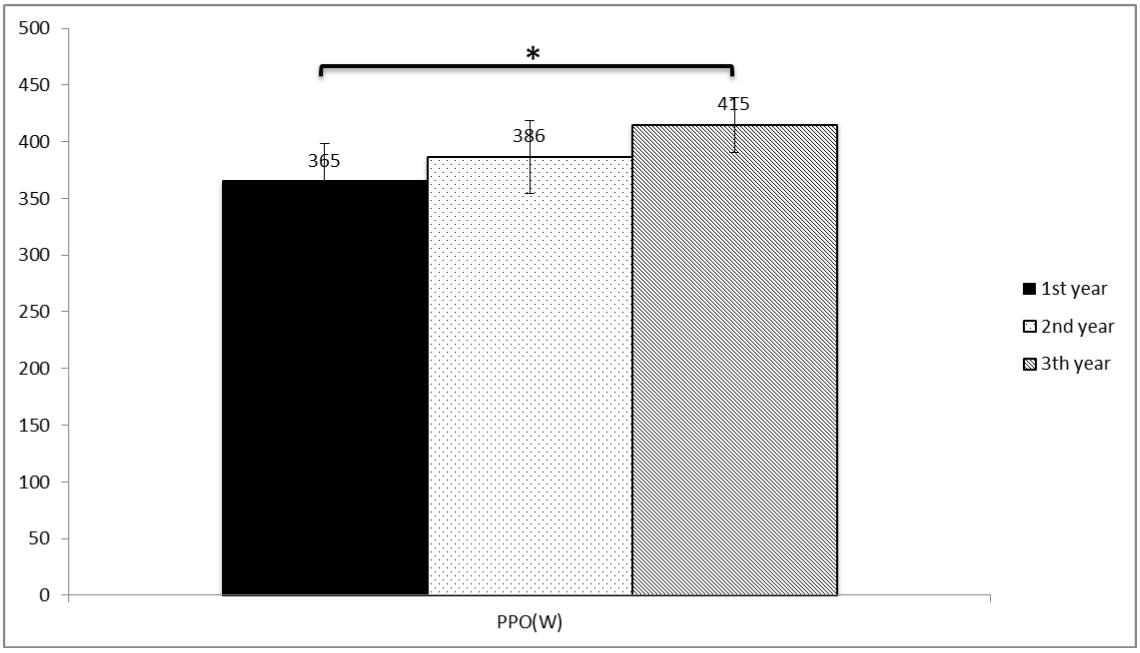


Figure 3: Means values of peak power output (PPO) over three years study period in world-class cyclists. Significant differences were found between 1st and 3th year ($p < 0.05$).

Why efficiency could change over time in cyclists? Three possibilities could be involved: 1) changes in the dead space (VD); 2) better mechanical performance of respiratory muscles; 3) changes in sensitivity of carotic bodies. Given that this study was retrospective, we could not choose variables to measure. But, not finding any change in VE/VCO_2 slope, we can assume that none of these variables had to change or at least have not conditioned changes in respiratory efficiency.

Concerning dead space, we know from experiments with goats (Mitchell, 1990) that ventilation is stimulated by anadded dead space and this seems to be so in healthy humans during exercise (Wood et al, 2008) resulting in an increased VE/VCO_2 slope, implying a lower ventilatory efficiency. However, as the exercise intensity increased, the effect of the added dead space was reduced (Babb et al, 2010). This effect of added space is considered a short term modulation (STM) (Babb et al, 2010), but there is no evidence to support ventilatory control during exercise being influenced by hyperpnoeic history (long term modulation, LTM) associated with dead-space loading in humans (Cathcart et al, 2005) and even less in high-level athletes.

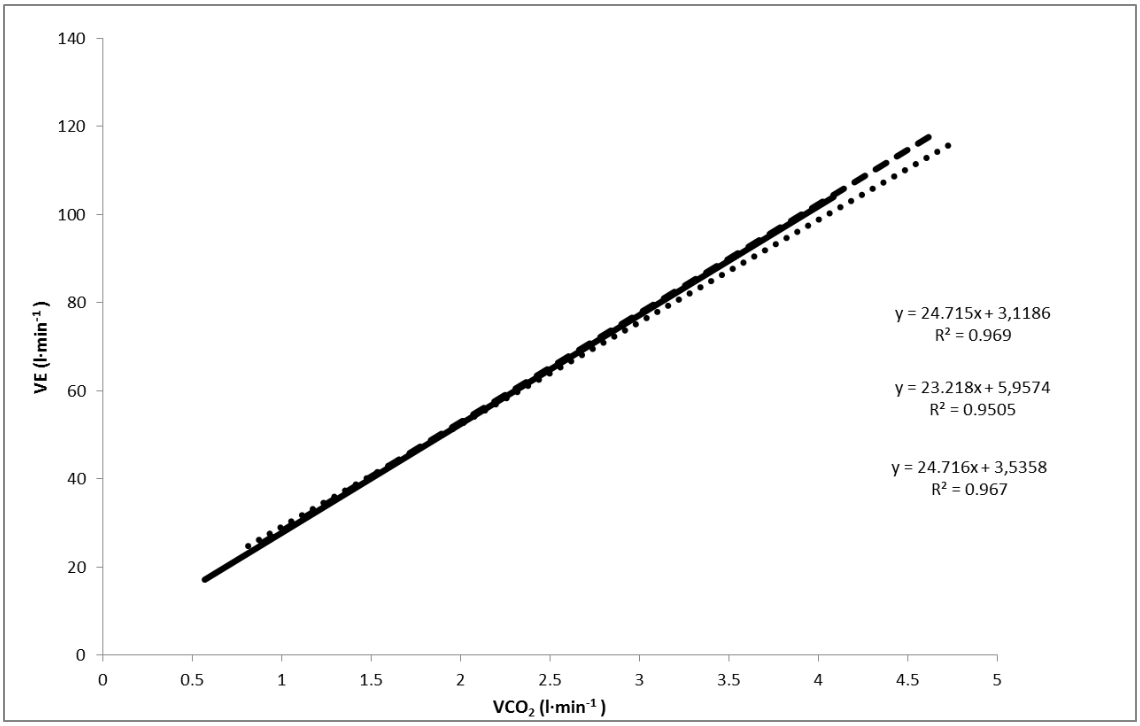


Figure 4: Relationship between ventilation (VE) and CO_2 output (VCO_2) showing values measured each year over the three-year observation period.

With respect to the mechanical performance of respiratory muscles, studies carried out with world-class professional cyclists, both transversal (Lucia et al., 2002) and longitudinal (Coyle, 2005; Hopker et al., 2010; Santalla et al., 2009; Sassi et al., 2008), found evidence that training increases muscular and mechanical performance. An inverse correlation between VO_{2max} and gross efficiency (GE) calculated as the ratio of work acomplished$\cdot$min^{-1} (i.e. W converted to Kcal$\cdot$min^{-1}), has been reported in world-class professional cyclists (Lucia et al., 2002). The authors suggested that this is a potential predictor of performance in professional cycling since the subjects with a higher GE were those with better results in time trials and in the overall standings in the Giro, Tour and Vuelta. In the same way, previous results obtained by our research group have described an increase in cycling efficiency in world-class professional cyclists, suggesting that this increase could even be a strategy to compensate a lower VO_{2max}, thereby maintaining a high competitive level (Santalla et al., 2009). Some studies suggest that specific respiratory muscle training can improve the endurance and strength of the respiratory muscles in healthy humans, although the effects on exercise performance remain controversial (Sheel, 2002). Therefore, it would be possible that a better trained diaphragm contributes to more efficient ventilation.

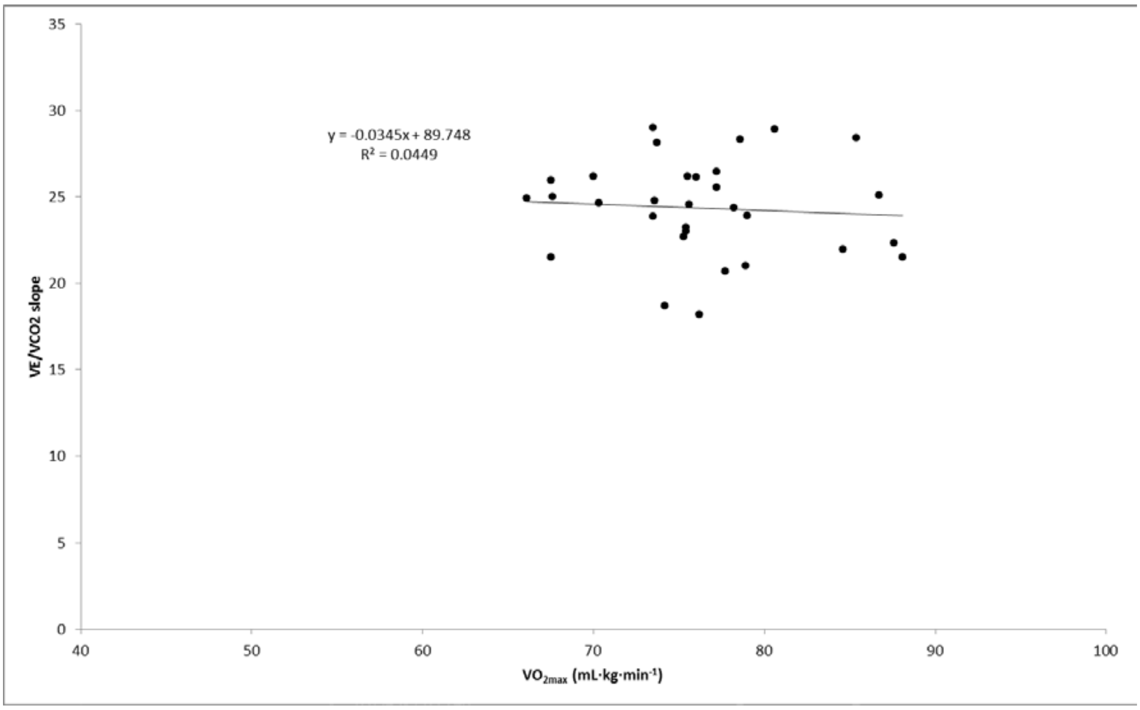


Figure 5: Relationship between ventilatory efficiency (VE/VCO_2 slope) and maximum oxygen uptake (VO_{2max}) over three years study period in world-class cyclists. No relationship was found between VE/VCO_2 slope and VO_{2max}.

Regarding sensitivity of chemoreceptors, we could not analyze it in a retrospective study, but we agree with other authors (Mitchel, 1990; Babb et al, 2010) that respiratory efficiency is not related to changes in chemoreflex stimulation.

Anyway, our results indicate that the training adaptations in high level athletes which cause changes in performance (Fig. 3) and other efficiency variables (Lucia et al., 2002; Santalla et al., 2009) do not modify ventilatory efficiency. The test-re-test reliability for VE/VCO_2 slope measurements was high (CCI=0.89), with a great coefficient of determination (all ≥ 0.8), this help to add evidence that the VE/VCO_2 slope remains unchanged over time. These subjects constantly increase their VE by approximately 24 $l \cdot min^{-1}$ for each $l \cdot min^{-1}$ of increase in VCO_2 during incremental cycling ergometry (Table 1). These values are indicative of high ventilatory efficiency, in fact they are lower than values found in juvenile cyclists (~28) (Brown et al., 2013).

Based on the few papers that have studied this variable during exercise, it seems that the VE/VCO_2 slope tends to remain constant, at least in healthy subjects (Sun et al., 2002). No significant differences in ventilatory efficiency have been found after 16-weeks of training (Brown et al., 2013), during exercise between sexes (Guerrero et al., 2008), age groups (Sun et al., 2002) or bodily position (Terkelsen et al., 1999).

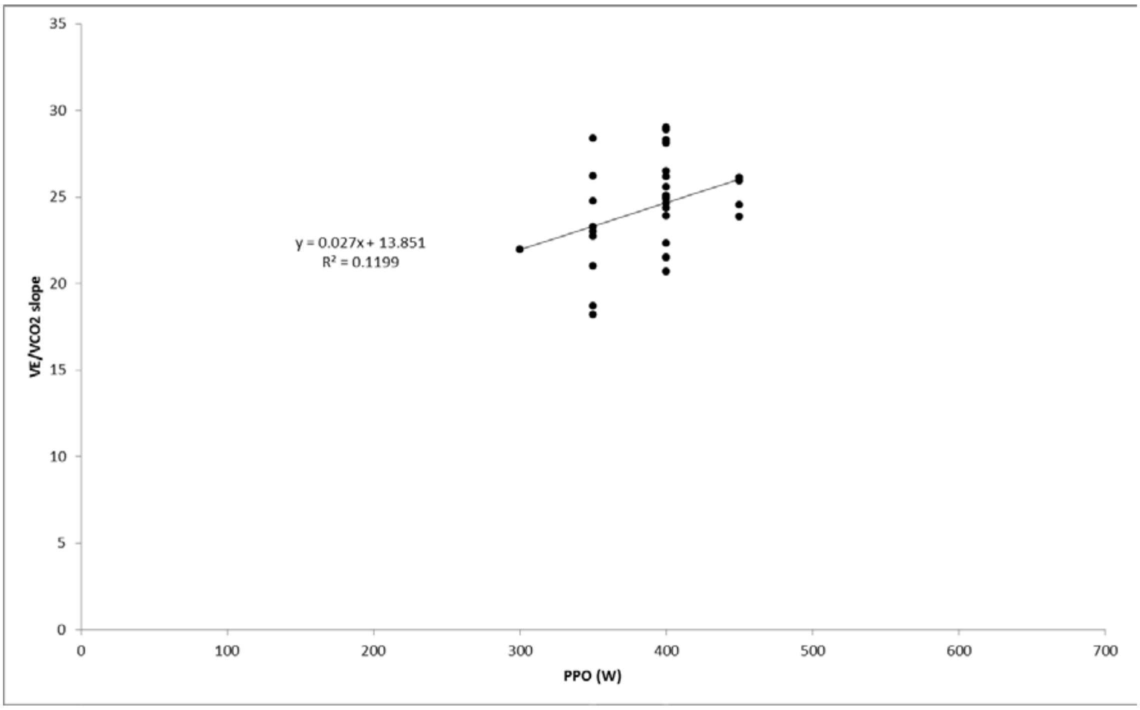


Figure 6: Relationship between ventilatory efficiency (VE/VCO_2 slope) and peak power output (PPO) over three years study period in world-class cyclists. No relationship was found between VE/VCO_2 slope and PPO.

Concerning the breathing pattern, both Vt/Ti and Ti/Ttot behave similarly in healthy people when ventilation is stimulated, regardless of the stimulus (exercise, CO_2 inhalation, etc.) (McConnell and Davies, 1992; Szekely et al., 1982).During progressive treadmill exercise, it is known thatTi/Ttot remained nearly constant while Vt/Ti increased linearly with VE and that this response was independent of gender and protocol (ramp or step) (Naranjo et al., 2005). However, only a few studies evaluated the ventilatory response to exercise in elite cyclists over time and most of them were limited to only analysis of changes in VE (Hoogeveen, 2000). Lucia et al. (1999) analyzed the breathing pattern in a cross-sectional study with highly competitive cyclists during incremental exercise, comparing amateur and professional cyclists. They demonstrated that Vt/Ti and Ti/Ttot showed a similar response in both groups. In addition, Scheuermann and Kowalchuk (1999) showed that Ti/Ttot and Vt/Ti were similar during a slow ramp (8 $W \cdot min^{-1}$) and fast ramp protocol (65 $W \cdot min^{-1}$) at all submaximal exercise intensities, suggesting that breathing pattern and respiratory timing may behave independently of alveolar and arterial PCO_2. Recently, Tanner et al. (2014) showed that there are no differences between cycling and running in breathing pattern variablesat maximal exercise intensities. There has been also reported an inter-individual variability in breathing patterns at high levels of exercise, suggesting that respiratory control by peripheral vagal afferents seems to prevail in subjects who increase f_R and not Vt after VT_2 (Gravier et al., 2013). In our study the breathing patter of athletes remain unchanged. Ti/Ttot values remained without significant change throughout the different tests (Table 1), while Vt/Ti values showed an almost perfect linear relationship with VE values (Figure 2). This indicates that the increases in VE during progressive exercise are associated with a proportional increase in the inspiratory driving activity without any alteration in the relationship between inspiration and expiration, even at the highest working intensities (over 400 W). Although this coincides with previous transversal evidence (Lucia et al., 1999; Lucia et al., 2001; Naranjo et al., 2005), it is the first time to be described longitudinally over a period of more than one season.

With respect to the variables related to performance, the stability of VO_{2max} observed (Fig. 2) over the three seasons and the increase in PPO (Fig. 3) agree with the previous research with world-class cyclists (Lucia et al., 2002; Santalla et al., 2009). Correlations analysis performed suggested that there is no relationship between the cycling performance and ventilatory efficiency in world-class cyclists (Fig. 5-6).

At least three limitations have to be addressed. First, we do not have exact data on the amount of training and competitions completed by the cyclists. However, due to the inclusion criteria, we strongly assume that all of them trained and competed similarly. Second, the sample size is relatively small but should be sufficiently large considering the known difficulties of gaining access to world-class cyclists. Thirdly, this study was retrospective and we could not choose the variables to measure; for example, we could not measure arterial PCO_2.

New research to evaluate the influence of specific training program on ventilatory efficiency and VA in athletes is necessary in order to better clarify the involvement and influence of this variable on ventilation performance during exercise. It would be useful to include measures of PaCO2 and VD in new designs.

5. Conclusions

In conclusion, the presented findings do not support our hypothesis that performance changes in world-class cyclists over a three-year period would be associated with changes in ventilatory efficiency and breathing pattern. They rather suggest that the central control of respiration, responsible for promoting efficient breathing patterns in response to exercise moves within very tight ranges, is set by the system and could not be modified through training and competition. Furthermore, the efficiency of the CO_2 elimination during exercise also appears to be preserved and closely related to the central control mechanism.

REFERENCES

Arena, R., Guazzi, M., Myers, J., 2007a.Prognostic value of end-tidal carbon dioxide during exercise testing in heart failure. Int. J. Cardiol. 117, 103-108.

Arena, R., Myers, J., Guazzi, M., 2008. The clinical and research applications of aerobic capacity and ventilatory efficiency in heart failure: an evidence-based review. Heart. Fail. Rev. 13, 245-269.

Arena, R., Myers, J., Hsu, L., Peberdy, M.A., Pinkstaff, S., Bensimhon, D., Chase, P., Vicenzi, M., Guazzi, M., 2007b. The minute ventilation/carbon dioxide production slope is prognostically superior to the oxygen uptake efficiency slope. J. Card. Fail. 13, 462-469.

Babb, T.G., Wood, H.E., Mitchell, G.S. 2010.Short- and long-term modulation of the exercise ventilatory response. Med. Sci. Sports Exerc. 42, 1681–1687.

Beltrão, B.A., Herdman, T.H., Pascoal, L.M., Chaves, D.B.R., Silva, V.M., Lopes, M.V., 2015. Ineffective breathing pattern in children and adolescents with congenital heart disease: accuracy of defining characteristics. J. Clin. Nurs. 24 (17-18), 2505-2513.

Benchetrit, G., 2000. Breathing pattern in humans: diversity and individuality.Respir. Physiol. 122, 123-129.

Brown, S.J., Raman, A., Schlader, Z., Stannard, S.R., 2013. Ventilatory efficiency in juvenile elite cyclists. J. Sci. Med. Sport 16, 266-270.

Cathcart AJ, Herrold N, Turner AP, Wilson J, Ward SA. 2005. Absence of long-term modulation of ventilation by dead-space loading during moderate exercise in humans. Eur. J. Appl. Physiol. 93, 411-420.

Coyle, E.F., 2005. Improved muscular efficiency displayed as Tour de France champion matures. J. Appl. Physiol. (Bethesda, Md. : 1985) 98, 2191-2196.

Fernandez-Garcia, B., Perez-Landaluce, J., Rodriguez-Alonso, M., Terrados, N., 2000.Intensity of exercise during road race pro-cycling competition.Med. Sci. Sports Exerc. 32, 1002-1006.

Gilbert, C., 2005. Better chemistry through breathing: The story of carbon dioxide and how it can go wrong. Biofeedback 33, 100-104.

Gravier, G., Delliaux, S., Delpierre, S., Guieu, R., Jammes, Y., 2013. Inter-individual differences in breathing pattern at high levels of incremental cycling exercise in healthy subjects. Respir. Physiol. Neurobiol. 189, 59-66.

Guerrero, L., Naranjo, J., Carranza, M.D., 2008. Influence of gender on ventilatory efficiency during exercise in young children. J. Sports Sci. 26, 1455-1457.

Hoogeveen, A.R., 2000. The effect of endurance training on the ventilatory response to exercise in elite cyclists. Eur. J. Appl. Physiol. 82, 45-51.

Hopker, J., Coleman, D., Passfield, L., Wiles, J., 2010. The effect of training volume and intensity on competitive cyclists' efficiency. Appl. Physiol. Nutr. Metab. 35, 17-22.

Ingle, L., Goode, K., Carroll, S., Sloan, R., Boyes, C., Cleland, J.G., Clark, A.L., 2007. Prognostic value of the VE/VCO2 slope calculated from different time intervals in patients with suspected heart failure. Int. J. Cardiol. 118, 350-355.

Laveneziana P, Agostoni P, Mignatti A, Mushtaq S, Colombo P, Sims D, Uriel N, Jorde UP. 2010. Effect of acute β-blocker withholding on ventilatory efficiency in patients with advanced chronic heart failure. J. Card. Fail. 16(7), 548-55.

Lucia, A., Carvajal, A., Calderon, F.J., Alfonso, A., Chicharro, J.L., 1999. Breathing pattern in highly competitive cyclists during incremental exercise. Eur. J. Appl. Physiol. Occup. Physiol. 79, 512-521.

Lucia, A., Hoyos, J., Pardo, J., Chicharro, J.L., 2001. Effects of endurance training on the breathing pattern of professional cyclists.Jpn. J. Physiol. 51, 133-141.

Lucia, A., Hoyos, J., Pérez, M., Santalla, A., Chicharro, J.L., 2002.Inverse relationship between VO2max and economy/efficiency in world-class cyclists.Med. Sci. Sports Exerc. 34, 2079-2084.

Lucia, A., Hoyos, J., Santalla, A., Earnest, C.P., Chicharro, J.L., 2003. Giro, Tour, and Vuelta in the same season. Br. J. Sports Med. 37, 457-459.

Lucia, A., Pardo, J., Durantez, A., Hoyos, J., Chicharro, J.L., 1998. Physiological differences between professional and elite road cyclists. Int. J. Sports Med. 19, 342-348.

McConnell, A.K., Davies, C.T., 1992. A comparison of the ventilatory responses to exercise of elderly and younger humans. J. Gerontol. 47, B137-141.

Milic-Emili, G., Cajani, F., 1957.[Frequency of breathing as a function of respiratory ventilation during rest].Boll. Soc. Ital. Biol. Sper. 33, 821-825.

Milic-Emili, J., 1982. Recent advances in clinical assessment of control of breathing. Lung. 160, 1-17.

Milic-Emili, J., Grunstein, M.M., 1976.Drive and timing components of ventilation.Chest. 70, 131-133.

Milsom, W.K., Abe, A.S., Andrade, D.V., Tattersall, G.J., 2004. Evolutionary trends in airway CO2/H+ chemoreception. Respir. Physiol. Neurobiol. 144, 191-202.

Mitchell GS, 1990.Ventilatory control during exercise with increased respiratory dead space in goats. J. Appl. Physiol. 69, 718-727.

Naranjo, J., Centeno, R.A., Galiano, D., Beaus, M., 2005.A nomogram for assessment of breathing patterns during treadmill exercise. Br. J. Sports Med. 39, 80-83.

Robertson, HT. 2011. Gas Exchange consequences of left heart failure. Compr. Physiol. 1, 621-634.

Sallet, P., Mathieu, R., Fenech, G., Baverel, G., 2006. Physiological differences of elite and professional road cyclists related to competition level and rider specialization. J. Sports Med. Phys. Fitness 46, 361-365.

Santalla, A., Naranjo, J., Terrados, N., 2009. Muscle efficiency improves over time in world-class cyclists. Med. Sci. Sports Exerc. 41, 1096-1101.

Sassi, A., Impellizzeri, F.M., Morelli, A., Menaspa, P., Rampinini, E., 2008. Seasonal changes in aerobic fitness indices in elite cyclists. Appl. Physiol. Nutr. Metab. 33, 735-742.

Scheuermann, B.W., Kowalchuk, J.M., 1999. Breathing patterns during slow and fast ramp exercise in man. Exp. Physiol. 84, 109-120.

Schneider, D.A., Berwick, J.P., 1998. VE and VCO2 remain tightly coupled during incremental cycling performed after a bout of high-intensity exercise. Eur. J. Appl. Physiol. Occup. Physiol. 77, 72-76.

Sheel AW. 2002. Respiratory muscle training in healthy individuals: physiological rationale and implications for exercise performance. Sports Med. 32, 567-581.

Sun, X.G., Hansen, J.E., Garatachea, N., Storer, T.W., Wasserman, K., 2002. Ventilatory efficiency during exercise in healthy subjects. Am. J. Respir. Crit. Care Med. 166, 1443-1448.

Szekely, O., Askanazi, J., Rosenbaum, S.H., Foster, R., Milic-Emili, J., Kinney, J.M., 1982. Ventilatory patterns during steady state and progressive exercise. Clin. Physiol. 2, 391-400.

Tanner, D.A., Duke, J.W., Stager, J.M., 2014. Ventilatory patterns differ between maximal running and cycling. Respir. Physiol. Neurobiol. 191, 9-16.

Terkelsen, K.E., Clark, A.L., Hillis, W.S., 1999. Ventilatory response to erect and supine exercise.Med. Sci. Sports Exerc. 31, 1429-1432.

Ukkonen, H., Burwash, I.G., Dafoe, W., de Kemp, R.A., Haddad, H., Yoshinaga, K., Davies, R.A., Gannon, E.K., Dasilva, J.N., Beanlands, R.S., 2008. Is ventilatory efficiency (VE/VCO(2) slope) associated with right ventricular oxidative metabolism in patients with congestive heart failure? Eur. J. Heart Fail. 10, 1117-1122.

Wood HE, Mitchell GS, Babb TG, 2008. Short-term modulation of the exercise ventilatory response in young men. J. Appl. Physiol. 104,244-52.

ESTUDIO II

INFLUENCE OF INSPIRATORY MUSCLE TRAINING ON VENTILATORY EFFICIENCY AND CYCLING PERFORMANCE IN NORMOXIA AND HYPOXIA

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INFLUENCE OF INSPIRATORY MUSCLE TRAINING ON VENTILATORY EFFICIENCY AND CYCLING PERFORMANCE IN NORMOXIA AND HYPOXIA

ABSTRACT

The aim of this study was to analyse the influence of inspiratory muscle training (IMT) on ventilatory efficiency, in normoxia and hypoxia, and to investigate the relationship between ventilatory efficiency and cycling performance. Sixteen sport students (23.05±4.7 years; 175.11±7.1 cm; 67.0±19.4 kg; 46.4±8.7 ml·kg-1·min-1) were randomly assigned to an inspiratory muscle training group (IMTG) and a control group (CG). The IMTG performed 2 training sessions/day (30 inspiratory breaths, 50% peak inspiratory pressure (Pimax), 5 days/wk, 6-wks. Before and after the training period subjects carried out an incremental exercise test to exhaustion with gas analysis, lung function testing and a cycling time trial test in hypoxia and normoxia. Simulated hypoxia (FiO2=16.45%), significantly altered the ventilatory efficiency response in all subjects ($p<0.05$). Pimax increased significantly in the IMTG whereas no changes occurred in the CG (time x group, $p<0.05$). Within group analyses showed that the IMTG improved ventilatory efficiency (VE/VCO2 slope; EqCO2VT2) in hypoxia ($p<0.05$) and cycling time trial performance (WTTmax(W); WTTmean(W); PTF(W)) ($p<0.05$) in hypoxia and normoxia. Significant correlations were not found in hypoxia nor normoxia found between ventilatory efficiency parameters (VE/VCO2 slope; LEqCO2; EqCO2VT2) and time trial performance. On the contrary the oxygen uptake efficiency slope (OUES) was highly correlated with cycling time trial performance ($r=0.89$; $r=0.82$; $p<0.001$) under both conditions. Even though no interaction effect was found, the within group analysis may suggest that IMT reduces the negative effects of hypoxia on ventilatory efficiency. In addition, the data suggest that OUES plays an important role in submaximal cycling performance.

Key words: VE/VCO2 slope / cycling performance / ventilation / chemosensitivity / time trial

1. Introduction

Ventilatory efficiency can be defined as the relationship between carbon dioxide production (VCO_2) and ventilation (V_E). Increased V_E and the removal of CO_2 during physical exercise are essential for homeostatic control of whole body pH (Brown et al., 2013). There are four common ways for measuring ventilatory efficiency during an incremental test: a) using the slope of the relationship between VCO_2 and V_E (V_E/VCO_2 slope) (Ingle et al., 2007), b) the lowest equivalent of CO_2 during the incremental test ($LEqCO_2$) (Sun et al., 2002), c) the equivalent of CO_2 at the second ventilatory threshold ($EqCO_2VT_2$) (Sun et al., 2002) and d) the oxygen uptake efficiency slope (OUES) (Baba et al., 1999a). Generally, a lower equivalent of CO_2 indicates a greater ventilatory efficiency (Sun et al., 2002). The OUES represents the rate of increase of VO_2 in response to a given V_E during incremental exercise, indicating how effectively oxygen is extracted and taken into the body (Baba et al., 1996).

In the clinic field ventilatory efficiency has been widely used as a prognostic marker to determine exercise limitation (Ingle et al., 2007;Arena et al., 2008;Laveneziana et al., 2010). Indeed, a relationship has been reported between sudden dead risk in hypertrophic cardiomyopathy and ventilatory efficiency (Magrì et al., 2016). However, the importance of the ventilatory efficiency for sport performance remains unclear. On one hand, Brown et al., (2013) did not find a relationship between maximum oxygen uptake (VO_{2max}) and OUES in juvenile cyclists. In the same way, we did not find a relationship between VO_{2max}, peak power output (PPO) and V_E/VCO_2 slope in world-class cyclists (Salazar-Martínez et al., 2016). On the other hand, a significant correlation was found between OUES and VO_{2max} in young active women (Mourot et al., 2004).

In hypoxia, the reduced partial pressure of oxygen (PO_2) and the resulting arterial desaturation stimulates V_E (Babcock et al., 1995). Although the increased V_E during exercise in hypoxia (FIO_2=0.15) increases PaO_2 (Warner and Mitchell, 1990) it also leads to a higher oxygen cost of breathing compared to normoxia (Babcock et al., 1995). Additionally, in hypoxia, V_E may increase in excess of what would be required to maintain partial pressure levels of carbon dioxide ($PaCO_2$) (Warner and Mitchell, 1990). Therefore, a certain ventilatory inefficiency could be expected in hypoxia due to this overshoot in V_E. However, high ventilatory efficiency is essential to maintain adequate level of PaO_2 and $PaCO_2$ with a lower breathing work in high altitude (Bernardi et al., 2006). In this regard, it may be assumed that efficient breathing may play an important role in the regulation of PaO_2 and $PaCO_2$ and achieving a higher sport performance in hypoxia.

In accordance, inspiratory muscle training (IMT) has been shown to be an effective method to improve both the ventilatory response in normoxia and hypoxia (Downey et al., 2007;Esposito et al., 2010) and the alveolar-arterial gradient in hypoxia (Esposito et al., 2010). Thus, it could be speculated that well trained inspiratory muscles may help to preserve PaO_2 in hypoxia due to improved ventilation-perfusion matching and to prevent excessive CO_2 output due to less hyperventilation. However, to the best of our knowledge, whether or not IMT may improve the ventilatory efficiency under hypoxia conditions has not yet been tested.

Next to the effect on ventilation, IMT has been shown to improve sport performance as well (Romer et al., 2002a;Wells et al., 2005). However, the mechanisms responsible for the performance improvements after IMT remain controversial (Edwards and Walker, 2009). The mechanisms suggested to improve performance include a hypertrophy of diaphragm (Downey et al., 2007), an increase in blood flow to the locomotor muscles (Harms et al., 1997) and a reduction in subjective perception of fatigue and dyspnea ratings (Downey et al., 2007). Additionally, Sheel (2002) hypothesised that changes in performance after IMT could be related to improvements on ventilatory efficiency. However, to the best of our knowledge there are no studies evaluating the relationship between changes in sport performance after IMT and ventilatory efficiency. After IMT, the metabolic demand of the inspiratory muscles during exercise are reduced (Babcock et al., 1995), thus contributing to a lower overall O_2 uptake and CO_2 output. In situations where the ventilatory efficiency is impaired, for example in hypoxia, such effects may influence exercise performance (Roussos, 1985).

Therefore, the aim of this study was a) to evaluate the influence of IMT on ventilatory efficiency in normoxia and hypoxia, and b) to investigate the relationship between ventilatory efficiency and cycling performance under both conditions.

We hypothesised that IMT improves ventilatory efficiency in normoxia and especially in hypoxia and reduces the metabolic demands of the respiratory muscles in both conditions. We also hypothesised that improvements in submaximal cycling performance can be linked to improvements in ventilatory efficiency in normoxia and hypoxia.

2. Materials and methods

2.1. Subjects

Sixteen physically active and healthy participants (n=9 male (23.44±2.7 years; 180.22±3.5 cm; 78.2±5.5 kg; 48.39±7.28 ml·kg^{-1}·min^{-1}); n=7 female (25.37±3.24 years; 168.75±5.1 cm; 62.62.2±9.47 kg; 38.15±6.57 ml·kg^{-1}·min^{-1}) were selected for the study. Each participant completed a health questionnaire before being included in the study. Participants with health diseases, breathing problems or obstructive defects were excluded from the study. Before starting the study, written informed consent was obtained from each participant in accordance with the Declaration of Helsinki. The study was approved by the Ethics Committee of the University of Innsbruck.

2.2. Design

Participants were randomly assigned to either an inspiratory muscle training group (IMTG) or a control group (CG). The IMTG performed 2 training sessions per day, 5 days per week during a period of 6 weeks. Each participant completed 30 inspiratory breaths with a PowerBreath device (PowerBreathe®, K3) at 50% of their individual Pimax. Inspiratory training load was adjusted weekly at 50% of the individual Pimax. Every training session was performed under expert supervision. The CG did not carry out any inspiratory training during the experimental period. This procedure seems to be adequate as a placebo effect is not expected. For instance, when considering differences between trials that included a control group and studies that did not, 69% of the placebo-controlled studies showed a positive outcome for RMT (i.e. performance improvements for the RMT groups significantly exceeded those for the control groups), which is very similar to the 75% positive outcomes of the studies without any controls (Illi et al., 2012). Participants were advised not to change normal physical training habits during the experimental period.

2.3. Pulmonary function tests

Before and after the experimental period, participants performed lung function testing (Schiller SP-1®, Switzerland) to assess the forced vital capacity (FVC), forced expiratory volume during the first second (FEV_1), the ratio between forced expiratory capacity during the first second and vital capacity (FEV_1/VC), the peak expiratory flow (PEF) and the peak inspiratory flow (PIF) (Table 1). The best attempt out of 3 tests was included in the analysis. Peak inspiratory mouth pressure (Pimax) was determined with a portable device (PowerBreathe®, K3). During the Pimax test participants had to inspire as fast as possible from a normal expiration. Each participant repeated the test until the measurements were stable. Pimax was measured weekly using the same testing protocol.

2.4. Incremental exercise testing

Before (Pre) and after (Post) the training period participants performed maximum incremental exercise tests in normoxia and hypoxia (overall 4 tests). Each test was separated by 48 hours. During the tests, oxygen uptake (VO_2), carbon dioxide output (VCO_2), respiratory exchange ratio (RER), ventilation (V_E), breathing frequency (BF), tidal volume (VT), oxygen equivalent ($EqVO_2$) and carbon dioxide equivalent ($EqCO_2$) were measured breath by breath with a portable gas analyser (Jaeger Oxygen™®, Germany). The system was calibrated prior to each test with gas mixtures of known concentration. Tests were carried out on a cycle ergometer (RBM Cyclus 2®, Germany). After 4 min of warming up, participants started the test at 50W and then the load was increased by 25W each minute until volitional exhaustion. Achievement of maximum oxygen uptake (VO_{2max}) was accepted when a plateau was found in the relationship between VO_2 and power output or when three of the four criteria for maximal VO_{2max} were obtained (Howley et al., 1995). Tests were carried out at approximately the same time of the day in an air-conditioned normobaric hypoxic chamber (size 4.75 x 2.25m, LowOxygen®, Germany). During the normoxia testing the hypoxic chamber was switched off whereas during the hypoxia setting the chamber was set at a simulated altitude of 2500m (FiO_2=16,45%). Participants were blinded to the simulated altitude of the hypoxic chamber. Participants were advised to avoid exhausting exercise one day before the tests and to take any ergogenic aids (e.g. caffeine).

2.5. Time trial performance

Ninety minutes after the incremental test, cycling endurance performance was evaluated by a 10 min time trial (TT). The cycle ergometer was shifted to a fixed pedal force in which power output was dependent on the pedalling rate. Pedal force for each participant was set in order that pedalling at 90 rpm produced 85% (rounded to 5 W) of peak power output determined by the incremental cycle ergometer test. During the test, cyclists were strongly encouraged to choose a maximal pedalling rate that could be maintained for the respective test duration. As with the incremental test, each participant performed the TT, under normoxic (TT_{nor}) and under hypoxic conditions

(TT_{hyp}) separated by 48 hours, before and after the experimental period. During each TT test peak power output (W_{max}), mean power output (W_{mean}) and pedal torque force (PTF) were recorded.

2.6. Test-retest reproducibly of time trial test

The coefficient of variation for the time trial test in the control group in normoxia was 14.9% in the pre-test and 15.9% in the post-test (Figure 1a). In hypoxia, the coefficient of variation for the time trial test in the control group was 17.9% in the pre-test and 17.8% in the post-test (Figure 1b). The intra-class correlation coefficient for the time trial test in the control group was 0.92 in normoxia and 0.93 in hypoxia.

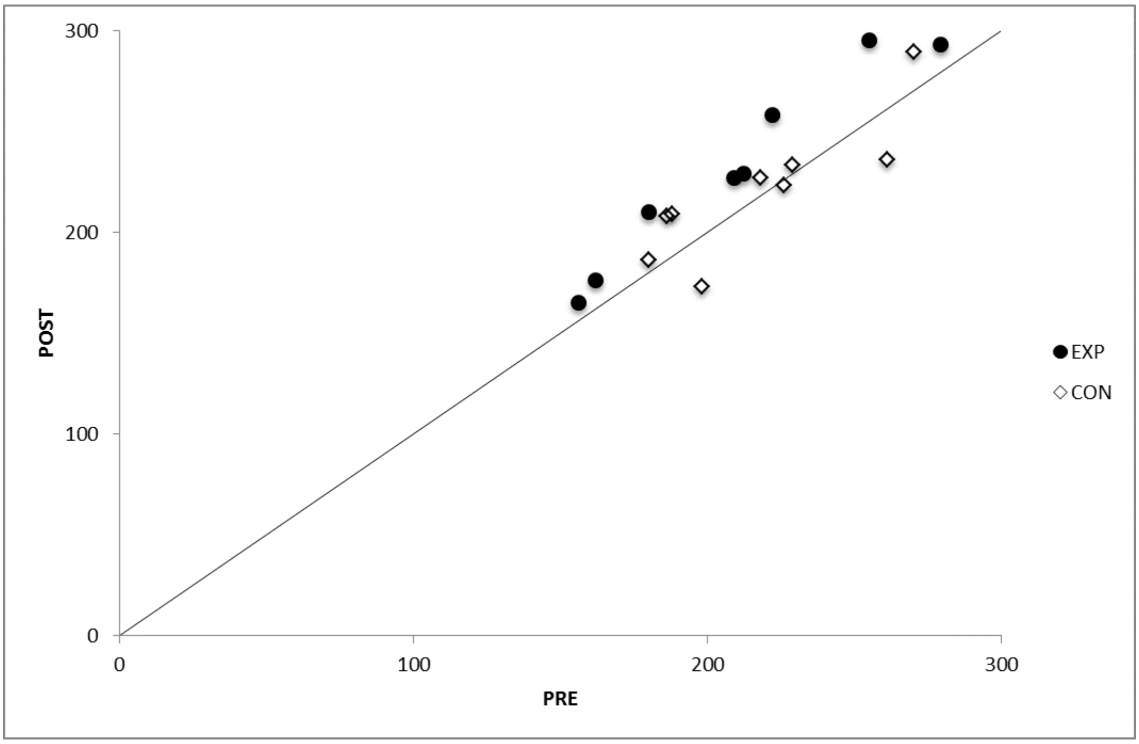

Figure 1a: Test-re-test reproducibility in the control/experimental group subjects during the time trial test (TT) test, before (Pre) and after (Post) the intervention period in normoxia. Identity lines are drawn in both graphs. See text for numerical analysis.

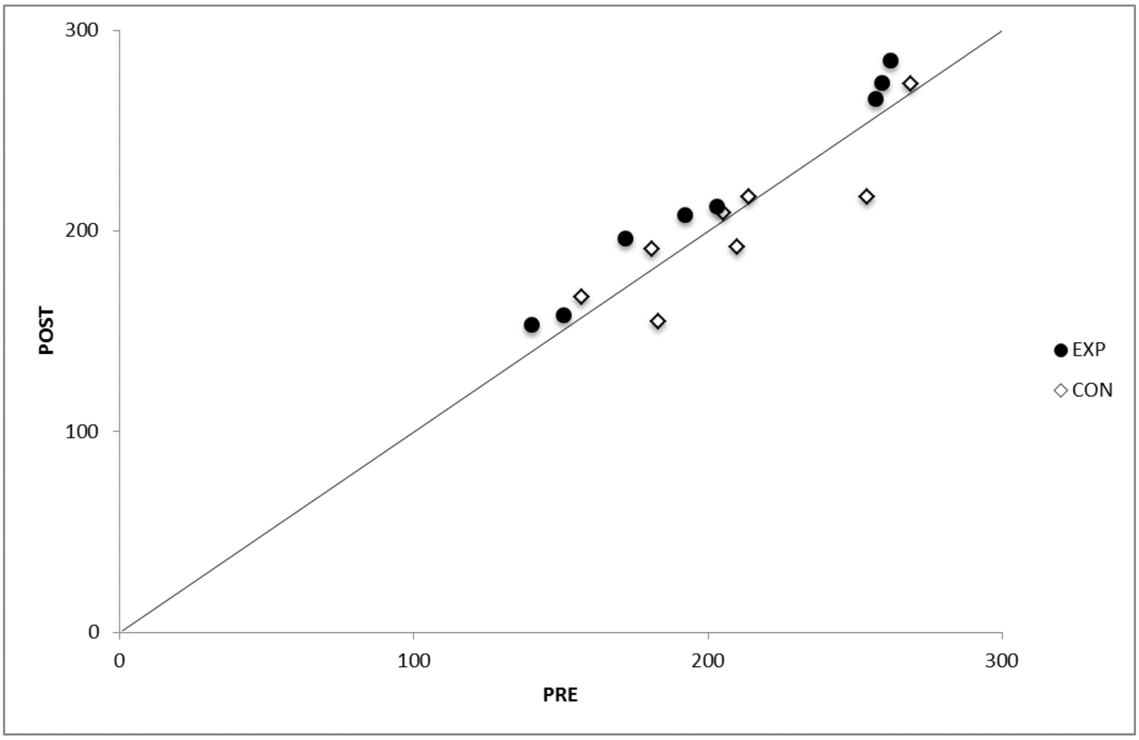

Figure 1b: Test-re-test reproducibility in the control/experimental group subjects during the time trial test (TT) test, before (Pre) and after (Post) the intervention period in hypoxia. Identity lines are drawn in both graphs. See text for numerical analysis.

2.7. Ventilatory efficiency

The V_E/VCO_2 slope was calculated from the slope of the relationship between VCO_2 and V_E during each incremental exercise test. To exclude the influence of the respiratory compensation due to acidosis during highly intensive exercise, the V_E/VCO_2 slope was determined from the beginning of the test until the second ventilatory threshold (VT_2). VT_2 was identified by an increase in the ventilatory equivalent of CO_2 ($EqCO_2$) and a decrease in end tidal partial pressure of carbon dioxide ($PETCO_2$) (Lucía et al., 2000). Oxygen uptake efficiency slope (OUES) was calculated from the linear relationship of VO_2 versus the logarithm of V_E during exercise ($VO_2 = a \log_{10} V_E + b$).

2.8. Statistics analysis

Data are expressed as mean ± SD for each variable. The statistical power for the chosen sample size of 16 participants (9 in the IMTG and 7 in the CG) was >90 %; alpha = 0.05. The power calculation (G*Power 3.1.7) was based on expected changes in Pimax and TT performance (W_{TTmean}) due to IMT (Romer et al., 2002a). The normal distribution of the data was checked by the Shapiro-Wilk test. The homogeneity of variance was evaluated by Levene´s test. To compare the values obtained for each variable during the test, mixed-effects ANOVA test was used (group x time x condition). When significant differences were found, the Bonferroni test was used as a post hoc test. ANOVA test was also applied to evaluate a possible gender effect (group x time x condition x gender). Effect size (ES) was calculated when a significant difference was found. A correlation analysis (Pearson-coefficient) was carried out between TT performance variables, incremental test variables and ventilatory efficiency variables with data from both groups and from both test in two different situations (normoxia and hypoxia) (Table 6). Linear regression analysis was performed between Pimax, V_E/VCO_2 slope and OUES (dependent variables) and TT performance (independent variable) with data from both groups (IMTG and CG) and from both tests (Pre and Post) in normoxia. The level of significance was set at $p<0.05$ for each statistical analysis.

3. Results

No gender effect was identified with regard to the parameters of interest (V_E/VCO_2 slope, $LEqCO_2$, $EqCO_2VT_2$, OUES). Baseline values did not differ between groups (V_E/VCO_2 slope, $LEqCO_2$, $EqCO_2VT_2$, OUES, Pimax, PPO, TT_{Wmean}). Outcomes of the pulmonary function testing before and after the experimental period are shown in Table 1. Significant differences were found in Pimax between Pre and Post-test in the IMTG ($p<0.05$).

Table 1: Results of pulmonary function testing Pre and Post experimental period (Mean±SD)

	IMTG		CG	
	Pre	Post	Pre	Post
FVC (l)	5.44±1.14	4.67±1.38	5.06±1.17	4.96±0.93
FEV_1 (l)	4.64±0.92	4.19±0.8	4,31±0.85	4.06±0.79
FEV_1/VC (%)	84.13±11.58	82.51±9.19	82.33±6.28	79.84±6.48
PEF ($l \cdot s^{-1}$)	9.27±2.23	8.2±1.53	8.9±2.47	8.73±2.4
PIF ($l \cdot s^{-1}$)	7.04±1.92	8.31±2.39	7.12±1.2	7.57±2.2
Pimax (cmH_2O)	119.6±37.36	166.91±42.65*	130.55±33.58	146.72±40.62

Forced vital capacity (FVC); forced expiratory volume during the first second (FEV_1); ratio between forced expiratory volume during the first second and vital capacity (FEV_1/VC); peak expiratory flow (PEF); peak inspiratory flow (PIF); peak inspiratory pressure (Pimax)

* $p<0.05$ post *vs* pre training

Table 2: Evaluation of ventilatory efficiency variables in normoxia and hypoxia before the experimental period with data from both groups (Mean±SD)

	V_E/VCO_2 slope	$LEqCO_2$	$EqCO_2VT_2$	OUES
Normoxia (n=16)	24.58±2.95	22.63±2.68	23.91±2.34	3.24±0.62
Hypoxia (n=16)	29.15±3.26*	24.8±1.9*	27.28±2.79*	2.96±0.85*
% ΔChange	+18.5%	+9.58%	+14.09%	-8.64%

Slope of the relationship between VCO_2 and V_E , (V_E/VCO_2 slope); lowest equivalent of CO_2 during the incremental test ($LEqCO_2$); equivalent of CO_2 in the second ventilatory threshold ($EqCO_2VT_2$); oxygen uptake efficiency slope (OUES).

* T-test for paired samples ($p<0.05$)

Percentage of change between measurements (% ΔChange)

3.1. Inspiratory muscle training (IMT)

Figure 2 shows the changes in Pimax during the experimental period in both groups. Mean Pimax improved significantly from week-1 to week-6 in the IMTG (+28.37%, $p<0.05$) with no improvements in the CG (interaction effect, time x group, $p<0.05$).

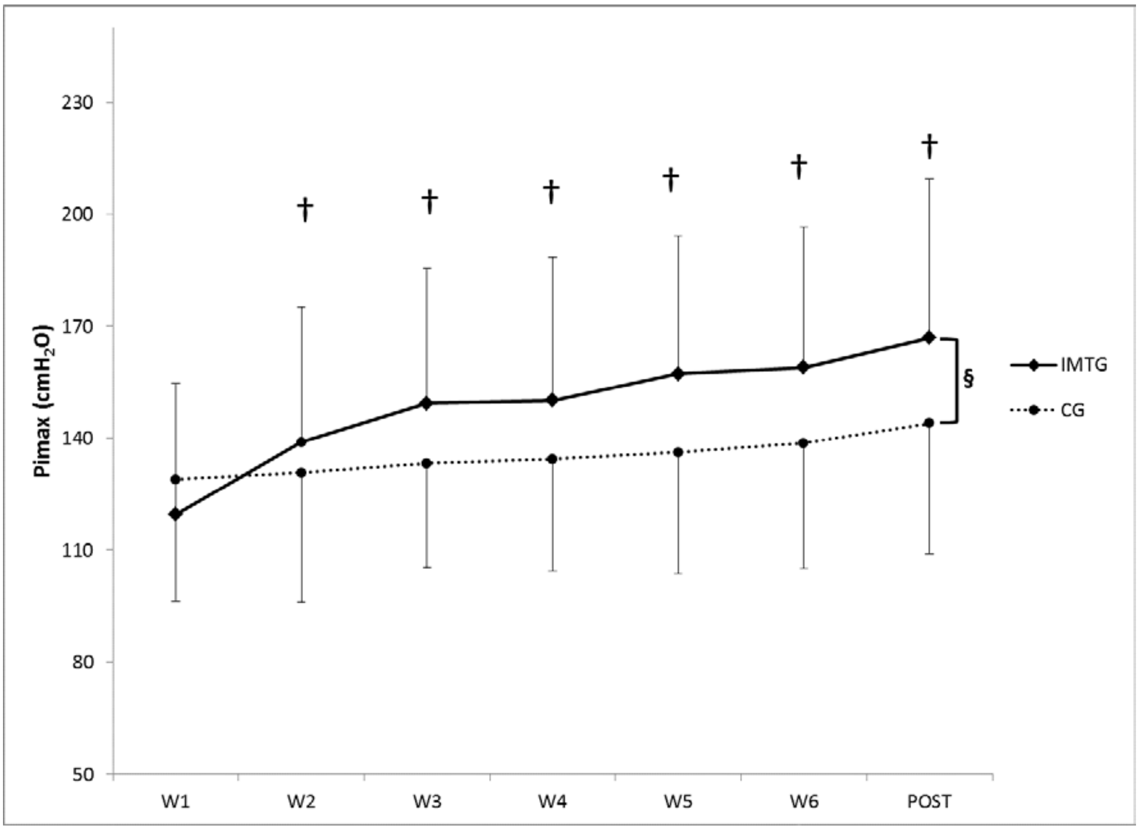

Figure 2: Weekly values of Pimax (Mean±SD) for the inspiratory muscle training group (IMTG) and control group (CG). § Two-way ANOVA for repeated measures (time x group) interaction ($p<0.05$). † Differences from baseline evaluation (Bonferroni test).

3.2. Ventilatory efficiency

The group comparison of the ventilatory efficiency variables are shown in Table 3. There were no group differences and no interaction effect in any of the established variables at Pre and Post in normoxia and hypoxia. Significant differences were found between normoxia and hypoxia in the V_E/VCO_2 slope, $LEqCO_2$ and $EqCO_2VT_2$ in the Pre-test in both groups ($p<0.05$) (Table 2). During the Post-test significant differences between normoxia and hypoxia were found in $LEqCO_2$ and OUES in the IMTG and in V_E/VCO_2 slope, $LEqCO_2$ and OUES in the CG ($p<0.05$). In both groups significant differences in V_E/VCO_2 slope and $EqCO_2VT_2$ were found between Pre and Post in hypoxia ($p<0.05$).

3.3. Time trial performance

Time trial performance parameters are shown in Table 4. During Pre and Post-test, significant differences between normoxia and hypoxia were found in $W_{TTmean\ (W)}$ and in $W_{TTmean\ (W/Kg)}$ in both groups ($p<0.05$). There was no interaction effect in these variables. However, after the experimental period, $W_{TTmean\ (W)}$ and $W_{TTmean\ (W/Kg)}$ were significantly higher in normoxia and hypoxia in the IMTG ($p<0.05$). At post, significant differences were found in W_{max} between normoxia and hypoxia in both groups ($p<0.05$). A significant reduction in PTF was found in the CG in hypoxia in both tests (Pre and Post) and in the Post-test in the IMTG ($p<0.05$). A significant increase in PTF was found in the IMTG in normoxia after IMT ($p<0.05$).

3.4. Incremental exercise testing

VO_{2max} was reduced in hypoxia in both groups compared to normoxia in the Post test ($p<0.05$). Compared to normoxia, PPO was reduced in both groups during the Pre and Post-test in hypoxia ($p<0.05$). There was no interaction effect. However, PPO increased significantly in the IMTG in normoxia after the experimental period compared to the Pre-test evaluation ($p<0.05$). Before the experimental period, V_{Emax} increased in hypoxic conditions in both groups. After the experimental period, V_{Emax} increased only in the CG. However, all these variations were not significant in both gorups. VT_{max} and BF_{max} did not change in any condition.

3.5. Correlation and regression analysis

Significant correlations were found between Pimax and W_{TTmean}, VO_{2max}, V_{Emax}, and PPO with data from both test and both groups in normoxia ($p<0.05$) (Table 6). No correlation was found between ventilatory efficiency variables and performance variables. A significant correlation was found between OUES and maximal performance variables (VO_{2max}, V_{Emax}, PPO) and W_{TTmean} ($p<0.05$) in normoxia and hypoxia (Table 6). A linear relationship was found between muscle breathing strength (Pimax) and TT performance in normoxia ($R^2=0.69$, $p=0.00$) (Figure 5) and in hypoxia ($R^2=0.67$, $p=0.00$). No relationship was found between time trial performance (W_{TTmean}) and ventilatory efficiency (V_E/VCO_2 slope) in normoxia ($R^2=0.149$, $p=0.02$) (Figure 5) and in hypoxia ($R^2=0.02$, $p=0.81$). W_{TTmean} and OUES were significantly related in normoxia ($R^2=0.647$, $p=0.00$) (Figure 5) and in hypoxia ($R^2=0.631$, $p=0.01$).

Table 3: Comparison between groups in ventilatory efficiency variables in the four experimental conditions. Data are presented as Mean±SD and Effect size (ES). ES is showed when a statistical difference was found.

	Pre		Post					
	Normoxia	Hypoxia	Normoxia	Hypoxia	ANOVA			
IMTG					Main Effect (time)	Main Effect (condition)	Main Effect (group)	Interaction (condition x group x time)
V_E/VCO_2 slope	23.68±2.94	28.77±2.74# (1.3)	25.6±3.95	26.48±2.77* (0.42)	0.267	0.000	0.476	0.313
$LEqCO_2$	22.51±1.32	24.72±1.45# (1.2)	22.73±1.65	23.98±1.64# (0.83)	0.609	0.000	0.755	0.519
$EqCO_2VT_2$	23.68±2.33	27.26±2.94# (1.3)	24.32±2.92	24.38±2.11* (1.5)	0.022	0.000	0.733	0.233
OUES	3.22±0.75	3.13±0.82	3.31±0.83	2.92±0.71# (0.81)	0.493	0.007	0.994	0.203
CG								
V_E/VCO_2 slope	25.31±2.35	29.81±3.7# (1.7)	25.63±3.93	28.06±3.5*# (1.2*-1.3#)				
$LEqCO_2$	22.75±3.59	24.87±2.17# (0.5)	22.62±1.98	24.73±1.58# (1.9)				
$EqCO_2VT_2$	24.26±2.05	27.15±2.81# (0.8)	24.15±3.11	25.7±2.73* (0.8)				
OUES	3.36±0.56	2.98±0.91	3.24±0.52	3.01±0.55# (1.0)				

Slope of the relationship between VCO_2 and V_E, (V_E/VCO_2 slope); lowest equivalent of CO_2 during the incremental test ($LEqCO_2$); equivalent of CO_2 in the second ventilatory threshold ($EqCO_2VT_2$); oxygen uptake efficiency slope (OUES). ANOVA mixed-effects Bonferroni post hoc test:

* Mixed-effects ANOVA Pre vs Post in the same condition ($p<0.05$)

Mixed-effects ANOVA Nor vs Hyp at the same time ($p<0.05$)

† Mixed-effects ANOVA IMTG vs CG ($p<0.05$)

Table 4: Comparison between groups in time trial variables in the four experimental conditions. Data are presented as Mean±SD and Effect size (ES). ES is showed when a statistical difference was found.

	Pre		Post					
	Normoxia	Hypoxia	Normoxia	Hypoxia	ANOVA			
IMTG					Main Effect (time)	Main Effect (condition)	Main Effect (group)	Interaction (condition x group x time)
$W_{TTmean\ (W)}$	217.25±49.07	204.5±49.67# (1.0)	241.87±56.01* (1.9)	219±51.22#* (0.6#-0.6*)	0.026	0.000	0.755	0.611
$W_{TTmean\ (W/Kg)}$	3.03±0.4	2.83±0.45# (1.3)	3.35±0.4* (2.4)	3.07±0.44#* (1.8#-1.5*)	0.041	0.000	0.823	0.610
$W_{TTmax\ (W)}$	296.25±109.6	282.3±112	319.12±118	289±105.9# (1.8)	0.969	0.000	0.353	0.769
$PTF_{\ (W)}$	147.5±22.83	141.25±27.35	156.87±29.51*(1.1)	144.37±27.57# (2.3)	0.466	0.000	0.676	0.164
CG								
$W_{TTmean\ (W)}$	221.25±32.5	209.12±37.47# (1.6)	222±35.25	202.62±36.23# (2.9)				
$W_{TTmean\ (W/Kg)}$	3.08±0.39	2.88±0.43# (1.6)	3.11±0.32	2.89±0.32# (2.8)				
$W_{TTmax\ (W)}$	273.28±28.87	258.37±36.89	265±39.84	238.7±37.45# (2.6)				
$PTF_{\ (W)}$	161.87±22.19	146.87±21.86# (1.7)	156.87±22.98	144.75±21.59# (1.0)				

Peak power output (W_{TTmax}); mean watts ($W_{TTmean\ (W)}$); mean watts per kilogram ($W_{TTmean\ (W/Kg)}$); pedal torque force (PTF). ANOVA mixed-effects Bonferroni post hoc test:

* Mixed-effects ANOVA Pre vs Post in the same condition ($p<0.05$)

\# Mixed-effects ANOVA Nor vs Hyp at the same time ($p<0.05$)

† Mixed-effects ANOVA IMTG vs CG ($p<0.05$)

Table 5: Measured cardiorespiratory and performance variables at maximal exercise intensity in the four experimental conditions. Data are presented as Mean±SD and Effect size (ES). ES is showed when a statistical difference was found.

	Pre		Post					
	Normoxia	Hypoxia	Normoxia	Hypoxia	ANOVA			
IMTG					Main Effect (time)	Main Effect (condition)	Main Effect (group)	Interaction (condition x group x time)
$VO_{2max\ (ml \cdot kg \cdot min^{-1})}$	47.19±9.45	45.15±7.34	45.86±5.07	43.37±6.88# (0.6)	0.139	0.018	0.973	0.731
$PPO_{\ (W)}$	289.37±55.12	274.62±53.28#(0.7)	306.62±58.86*(0.9)	281.5±51.37# (2.0)	0.180	0.000	0.778	0.660
$\dot{V}_{Emax\ (l \cdot min^{-1})}$	141.12±32.24	146.75±34.58	150.37±28.99	143.62±23.46	0.785	0.525	0.919	0.105
$VT_{max\ (l)}$	3.06±0.79	3.07±0.72	3.04±0.58	3.03±0.65	0.779	0.911	0.628	0.865
$BF_{max\ (breaths \cdot min^{-1})}$	57.25±5.54	56.5±7.72	57.12±5.93	56±7.38	0.468	0.711	0.488	0.850
CG								
$VO_{2max\ (ml \cdot kg \cdot min^{-1})}$	49±8.37	43.78±7.24	46.51±4.1	42.67±4.06# (2.3)				
$PPO_{\ (W)}$	306.62±41.86	285.5±43.1# (2.1)	307.75±47.67	280.25±44.65# (1.9)				
$\dot{V}_{Emax\ (l \cdot min^{-1})}$	143.5±35.97	148.5±42.57	135.87±40.92	147.12±43.6				
$VT_{max\ (l)}$	2.87±0.75	2.93±0.77	2.88±0.8	2.87±0.82				
$BF_{max\ (breaths \cdot min^{-1})}$	59.25±10.87	62±9.81	57.75±14	59.12±11.24				

Maximum oxygen uptake (VO_{2max}); peak power output (PPO); maximum ventilation ($\dot{V}_{Emax}$); maximum tidal volume (VT_{max}); maximum breathing frequency (BF_{max}). ANOVA mixed-effects Bonferroni post hoc test:

* Mixed-effects ANOVA Pre *vs* Post in the same condition ($p<0.05$)

Mixed-effects ANOVA Nor *vs* Hyp at the same time ($p<0.05$)

† Mixed-effects ANOVA IMTG *vs* CG ($p<0.05$)

Table 6: Correlation analysis between performance variables and ventilatory efficiency variables after experimental protocol with data from both groups.

	Normoxia				
	Pearson-r				
	$W_{TTmean\ (W)}$	$VO_{2max\ (ml\cdot kg\cdot min^{-1})}$	$V_{Emax\ (l\cdot min^{-1})}$	$Pi_{max\ (cmH2O)}$	PPO $_{(W)}$
Pimax $_{(cmH2O)}$	0.607*	0.503*	0.859*	1	0.623*
V_E/VCO_2 slope	0.126	0.153	0.278	0.361	0.036
$LEqCO_2$	0.011	0.083	0.288	0.274	-0.064
$EqCO_2VT_2$	-0.1	0.026	0.062	0.196	-0.220
OUES	0.89*	0.683*	0.669*	0.454	0.913*
	Hypoxia				
	Pearson-r				
	$W_{TTmean\ (W)}$	$VO_{2max\ (ml\cdot kg\cdot min^{-1})}$	$V_{Emax\ (l\cdot min^{-1})}$	$Pi_{max\ (cmH2O)}$	PPO $_{(W)}$
Pimax $_{(cmH2O)}$	0.599*	0.477	0.545*	1	0.587*
V_E/VCO_2 slope	0.060	0.045	0.145	0.304	0.029
$LEqCO_2$	-0.250	-0.084	-0.029	0.069	-0.283
$EqCO_2VT_2$	-0.105	-0.016	-0.019	0.131	-0.166
OUES	0.828*	0.664*	0.79*	0.408	0.885*

Peak inspiratory pressure (Pimax); Slope of the relationship between VCO_2 and V_E , (V_E/VCO_2 slope); lowest equivalent of CO_2 during the incremental test ($LEqCO_2$); equivalent of CO_2 in the second ventilatory threshold ($EqCO_2VT_2$); oxygen uptake efficiency slope (OUES); Maximum oxygen uptake (VO_{2max}); peak power output (PPO); maximum ventilation (V_{Emax}); mean watts (W_{TTmean}).
*Significant correlation ($p<0.05$)

4. Discussion

To the best of our knowledge, this is the first study that investigated the effects of inspiratory muscle training (IMT) on ventilatory efficiency variables in normoxic and hypoxic conditions. We hypothesised that IMT could improve the ventilatory efficiency response in normoxia and hypoxia. We also hypothesised that improvements in the submaximal cycling performance may be linked to improvements in ventilatory efficiency. The main finding of this study was that IMT improved V_E/VCO_2 slope (-7.95%) in hypoxia and TT performance in both normoxia (+10.17%) and hypoxia (+6.62%) conditions. However, despite this within-group effect no interaction effect was found. Additionally, cycling TT performance was positively related to the oxygen uptake efficiency slope (OUES) in normoxia and hypoxia and to the inspiratory muscle strength (Pimax). These findings partly support (only in OUES) the hypothesis that changes in sport performance after IMT may be linked to changes in ventilatory efficiency.

It is well known that V_E is stimulated in hypoxia mediated by stimulation of the peripheral chemoreceptors (Dempsey and Forster, 1982;Townsend et al., 2002). However, the influence of hypoxic conditions on ventilatory efficiency has not been thoroughly investigated. In the present study, hypoxia at Pre worsened the ventilatory efficiency response in both groups (+18.5% V_E/VCO_2 slope; +9.58% $LEqCO_2$; +14.09% $EqCO_2VT_2$ and -8.64% OUES, respectively) (Table 2), which is in agreement with the increase in the ventilatory equivalents described at 16% FiO_2 (Ozcelik and Kelestimur, 2004). The increased V_E at altitude, initiated to maintain SaO_2 (Rusko et al., 2004;Burtscher et al., 2006;Faiss et al., 2014), may to some extent explain the deterioration of ventilatory efficiency. In addition, in hypoxia V_E may be increased in excess of what would be required to maintain partial pressure levels of carbon dioxide ($PaCO_2$) (Warner and Mitchell, 1990) thus influencing the relationship between V_E and VCO_2.

After the training period, hypoxia did not significantly increase the V_E/VCO_2 slope response in the subjects that performed the IMT (+3.43% (ES=0.4) vs. +9.48% (ES=1.3) for the IMTG and CG, respectively). These changes might be explained by improvements in breathing muscle strength (Pimax) and altered breathing patterns after IMT. Respiratory muscle training has been shown as an effective method to improve A-a gradient and ventilation-perfusion mismatch (Esposito et al., 2010). A better A-a gradient in hypoxia might have reduced the V_E overshoot observed in hypoxia before training (Table 3). In support of this, it has been reported that climbers who managed to climb Mt. Everest and K2 without oxygen, are those with a high ventilatory efficiency and "optimized" breathing patterns (Bernardi et al., 2006). Again, it has to be underlined that no interaction effect existed with respect to the altered V_E/VCO_2 slope in hypoxia after the training period. Therefore, the reported training effect contains some uncertainty and further studies with a greater sample size are needed to confirm our conclusions.

With regard to cycling performance, TT performance was reduced significantly in both groups before IMT in hypoxia (Table 4). After IMT, only the IMTG improved TT performance in normoxia and hypoxia (Table 4). Our results support previous studies showing a positive effect of IMT on sport performance (Volianitis et al., 2001;Romer et al., 2002b;a;Edwards and Walker, 2009). However, the participants of the present study not only improved their performance in normoxia (+11.33%), they also improved their performance in hypoxia (+7.33%) despite a reduction in VO_{2max} (-5.42%) (Table 4). It could be hypothesised that IMT reduced the oxygen cost of the breathing muscles allowing higher O_2 availability for the locomotor muscles. In addition, it has been suggested that reductions in respiratory effort could lead to greater locomotor muscle recruitment mediated by central nervous system control (Edwards and Walker, 2009). Once more, it should be noted that despite the improvements found in the IMTG no interaction effect was found. Thus, outcomes should be interpreted with some caution.

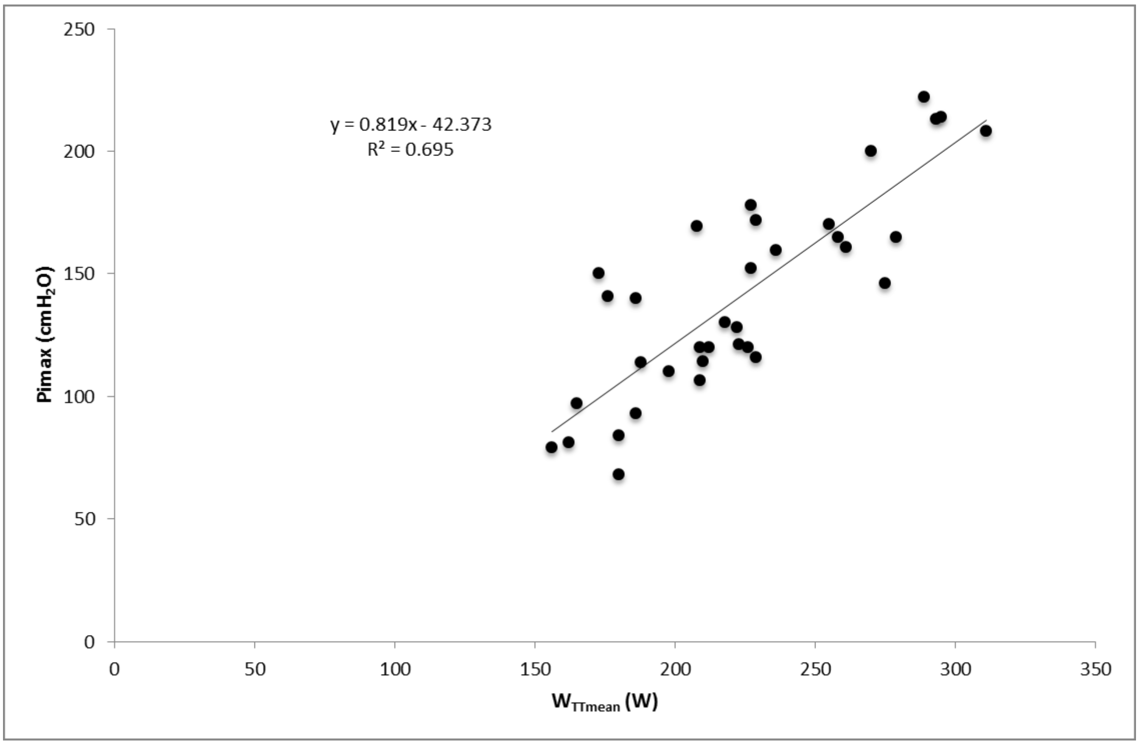

Figure 3: Relationship between inspiratory muscle strength (Pimax) and cycling time trial (TT) performance with data from both groups and both test in normoxia.

A further finding of the present investigation was that most of the ventilatory efficiency variables (V_E/VCO_2 slope, $LEqCO_2$ and $EqCO_2VT_2$) were not related to TT performance (Table 6) (Figure 5). This is in contrast to the finding of Sheel (2002) who suggests that improvements in submaximal exercise performance after IMT are related to improvements in ventilatory efficiency. Nonetheless, results of the present investigation show a positive relationship between cycling time trial performance and respiratory muscle strength (r^2=0.695) (Figure 5). It could be argued that the increased respiratory muscle strength might reduce the oxygen cost of breathing during submaximal exercise, thus improving oxygen delivery to the working limb muscles. However, further studies are necessary to confirm this hypothesis. In contrast, OUES showed a linear relationship with cycling TT performance (r^2=0.647) (Figure 5). Subjects who showed a lower oxygen cost for the same increment in V_E are those who achieved a higher performance in the TT (W_{TTmean}) (Table 6). However, OUES was not modified by the IMT (Table 3) and was not related to Pimax (Table 6). Therefore, IMT seems to not play a role in this relationship. It should be mentioned that in contrast to our trained sample, OUES was modified by IMT in patients with heart failure and weakened breathing muscle (Winkelmann et al., 2009).

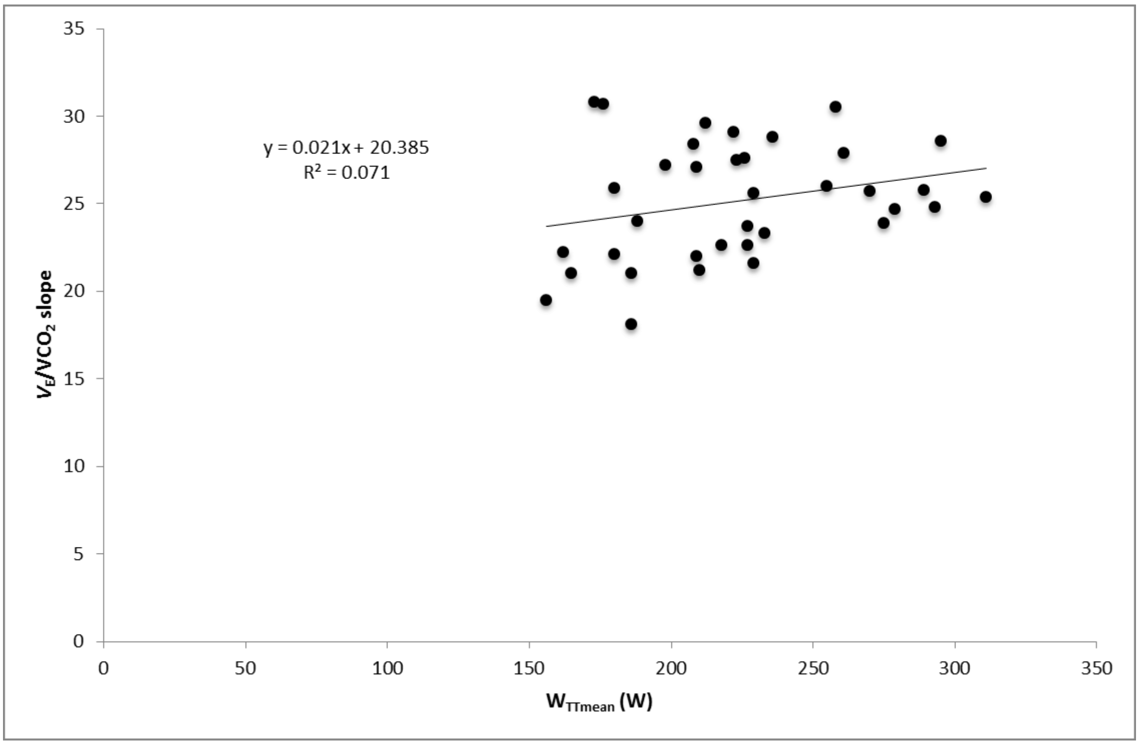

Figure 4: Relationship between cycling time trial (TT) performance and ventilatory efficiency measured as V_E/VCO_2 slope with data from both groups and both test in normoxia.

Regarding the incremental exercise test, VO_{2max} in hypoxia was reduced in both groups (-8,99% IMTG; -11.92% CG). Similar reductions were found previously at this simulated altitude (Lawler et al., 1988;Martin and O'Kroy, 1993). Additionally, IMT did not improve VO_{2max} in normoxia and hypoxia. Our results support previous studies that did not find an effect of IMT on VO_{2max} in normoxia and hypoxia (Downey et al., 2007;Esposito et al., 2010). However, the IMTG improved PPO after IMT in normoxia (+5.62%) and hypoxia (+2.51%) which is in contrast to previous studies that reported only a slight influence of IMT on PPO (Sheel, 2002;Illi et al., 2012). Moreover, except for OUES, the ventilatory efficiency variables were not correlated with performance variables of the incremental test (Table 6). Similar to the time trial outcome, OUES showed a strong correlation with VO_{2max} in normoxia and hypoxia (r=0.89 and r=0.82; respectively) and with PPO (r=0.91 and r=0.88; respectively). With respect to this finding, there are contrasting results reported in the literature. There are studies reporting a correlation between VO_{2max} and OUES (Baba et al., 1999a;Baba et al., 1999b;Baba et al., 1999c;Hollenberg and Tager, 2000) and others that did not find a correlation between these two parameters or only a weak correlation (Brown, 2010;Brown et al., 2013). Further research is necessary on the influence of IMT on ventilatory efficiency parameters in hypoxia.

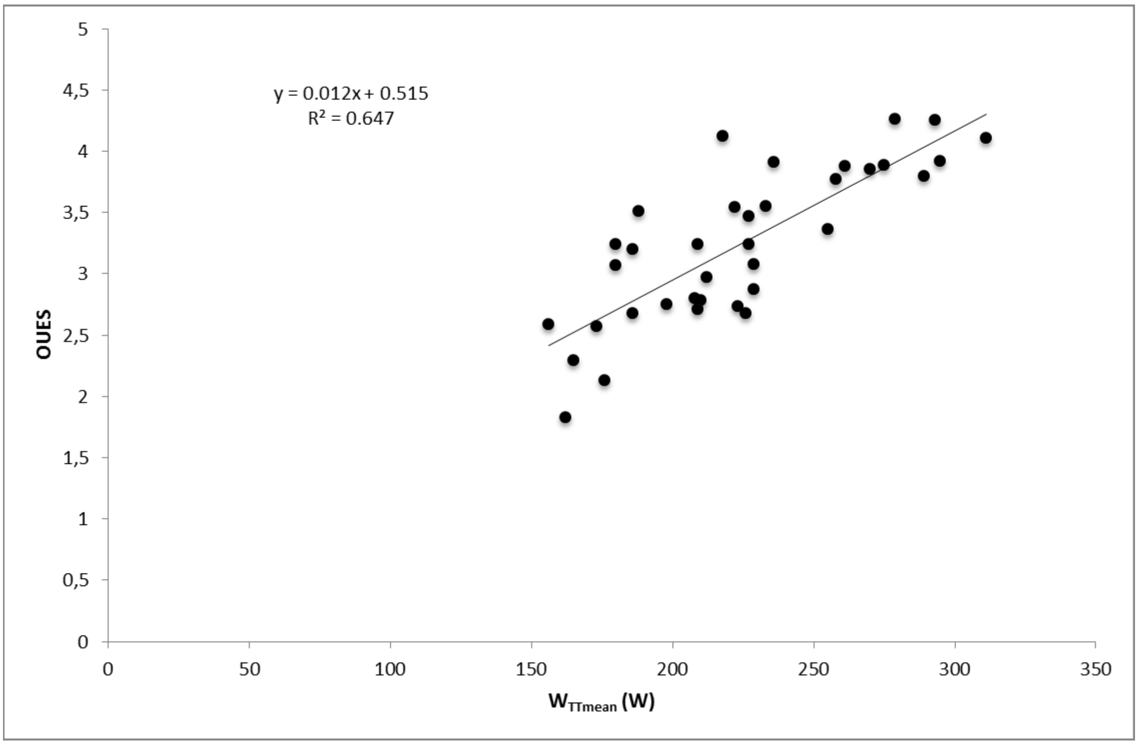

Figure 5: Relationship between cycling time trial (TT) performance and ventilatory efficiency measured as OUES with data from both groups and both test in normoxia.

Some limitations have to be addressed. First, the sample size was large enough to detect changes in Pimax and performance within the intervention group but might have been too low to detect group differences. Second, we do not have exact data on the amount of training and competitions completed by the subjects apart from the inspiratory training load. However, participants were advised not to change their usual training habits during the experimental period. All participants were enrolled in the same practical courses, and all reported to only have limited time for sports outside of the university setting. In addition, it was reported that conventional training (no specific breathing training) does not improve breathing muscle strength (Illi et al., 2012). Therefore, we can assume that they completed approximately the same training apart from the IMT and this did not influence our results. Third, we did not measure the oxygen saturation (SaO_2) during the hypoxia trials and this may have contributed important information. Lastly, we did not control for a possible placebo effect. However, as it is stated in the methods section, a large placebo effect is not expected. Therefore, we are confident that our conclusions were not affected.

5. Conclusions

Even though sample size might have been too low to show an interaction effect, the results of the present study suggest a possible positive effect of inspiratory muscle training on cycling time trial performance in both normoxic and hypoxic conditions. Additionally, this study shows that hypoxia has a negative effect on the ventilatory efficiency and that IMT may reduce this effect. Finally, the data suggest that except or OUES, ventilatory efficiency measures seem not to affect cycling time trial performance. These findings may have relevance for athletes planning to complete a high altitude training camp or for athletes competing at high altitude. Inspiratory muscle training before a competition at altitude might be a successful method to improve performance.

6. Authors contribution

Conception and design of the experiments: ES, AS and JNO; pre-testing, experimental preparation, data collection and analysis: ES, HG. The first version of the manuscript was written by ES, HG, MB, JNO and AS. All co-authors read and approved the final version of the manuscript.

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REFERENCES

Arena, R., Myers, J., and Guazzi, M. (2008). The clinical and research applications of aerobic capacity and ventilatory efficiency in heart failure: an evidence-based review. *Heart Fail. Rev.* 13**,** 245-269.

Baba, R., Kubo, N., Morotome, Y., and Iwagaki, S. (1999a). Reproducibility of the oxygen uptake efficiency slope in normal healthy subjects. *J. Sports Med. Phys. Fitness* 39**,** 202.

Baba, R., Nagashima, M., Goto, M., Nagano, Y., Yokota, M., Tauchi, N., and Nishibata, K. (1996). Oxygen intake efficiency slope: a new index of cardiorespiratory functional reserve derived from the relationship between oxygen consumption and minute ventilation during incremental exercise. *J. Am. Coll. Cardiol.* 59**,** 55-62.

Baba, R., Nagashima, M., Nagano, Y., Ikoma, M., and Nishibata, K. (1999b). Role of the oxygen uptake efficiency slope in evaluating exercise tolerance. *Arch. Dis. Child.* 81**,** 73-75.

Baba, R., Tsuyuki, K., Kimura, Y., Ninomiya, K., Aihara, M., Ebine, K., Tauchi, N., Nishibata, K., and Nagashima, M. (1999c). Oxygen uptake efficiency slope as a useful measure of cardiorespiratory functional reserve in adult cardiac patients. *Eur. J. Appl. Physiol. Occup. Physiol.* 80**,** 397-401.

Babcock, M.A., Johnson, B.D., Pegelow, D.F., Suman, O.E., Griffin, D., and Dempsey, J.A. (1995). Hypoxic effects on exercise-induced diaphragmatic fatigue in normal healthy humans. *J. Appl. Physiol.* 78**,** 82-92.

Bernardi, L., Schneider, A., Pomidori, L., Paolucci, E., and Cogo, A. (2006). Hypoxic ventilatory response in successful extreme altitude climbers. *Eur. Respir. J.* 27**,** 165-171.

Brown, S.J. (2010). Cardio-respiratory system efficiency in trained endurance cyclists. *Med. Sportiva* 14**,** 176-181.

Brown, S.J., Raman, A., Schlader, Z., and Stannard, S.R. (2013). Ventilatory efficiency in juvenile elite cyclists. *J. Sci. Med. Sport* 16**,** 266-270.

Burtscher, M., Faulhaber, M., Flatz, M., Likar, R., and Nachbauer, W. (2006). Effects of short-term acclimatization to altitude (3200 m) on aerobic and anaerobic exercise performance. *Int. J. Sports Med.* 27**,** 629-635.

Dempsey, J.A., and Forster, H.V. (1982). Mediation of ventilatory adaptations. *Physiol. Rev.* 62**,** 262-346.

Downey, A.E., Chenoweth, L.M., Townsend, D.K., Ranum, J.D., Ferguson, C.S., and Harms, C.A. (2007). Effects of inspiratory muscle training on exercise responses in normoxia and hypoxia. *Respir. Physiol. Neurobiol.* 156**,** 137-146.

Edwards, A.M., and Walker, R.E. (2009). Inspiratory muscle training and endurance: a central metabolic control perspective. *Int. J. Sports Physiol. Perform.* 4**,** 122-128.

Esposito, F., Limonta, E., Alberti, G., Veicsteinas, A., and Ferretti, G. (2010). Effect of respiratory muscle training on maximum aerobic power in normoxia and hypoxia. *Respir. Physiol. Neurobiol.* 170**,** 268-272.

Faiss, R., Von Orelli, C., Deriaz, O., and Millet, G.P. (2014). Responses to exercise in normobaric hypoxia: comparison of elite and recreational ski mountaineers. *Int. J. Sports Physiol. Perform.* 9.

Harms, C.A., Babcock, M.A., Mcclaran, S.R., Pegelow, D.F., Nickele, G.A., Nelson, W.B., and Dempsey, J.A. (1997). Respiratory muscle work compromises leg blood flow during maximal exercise. *J. Appl. Physiol.* 82**,** 1573-1583.

Hollenberg, M., and Tager, I.B. (2000). Oxygen uptake efficiency slope: an index of exercise performance and cardiopulmonary reserve requiring only submaximal exercise. *J. Am. Coll. Cardiol.* 36**,** 194-201.

Howley, E.T., Bassett, D.R., and Welch, H.G. (1995). Criteria for maximal oxygen uptake: review and commentary. *Med. Sci. Sports Exerc.* 27**,** 1292-1301.

Illi, S.K., Held, U., Frank, I., and Spengler, C.M. (2012). Effect of respiratory muscle training on exercise performance in healthy individuals. *Sports Med.* 42**,** 707-724.

Ingle, L., Goode, K., Carroll, S., Sloan, R., Boyes, C., Cleland, J.G.F., and Clark, A.L. (2007). Prognostic value of the V E/V CO2 slope calculated from different time intervals in patients with suspected heart failure. *Int. J. Cardiol* .118**,** 350-355.

Laveneziana, P., Agostoni, P., Mignatti, A., Mushtaq, S., Colombo, P., Sims, D., Uriel, N., and Jorde, U.P. (2010). Effect of Acute β-blocker Withholding on Ventilatory Efficiency in Patients With Advanced Chronic Heart Failure. *J. Card. Fail.* 16**,** 548-555.

Lawler, J., Powers, S.K., and Thompson, D. (1988). Linear relationship between VO2max and VO2max decrement during exposure to acute hypoxia. *J. Appl. Physiol.* 64**,** 1486-1492.

Lucía, A., Hoyos, J., Pérez, M., and Chicharro, J.L. (2000). Heart rate and performance parameters in elite cyclists: a longitudinal study. *Med. Sci. Sports Exerc.* 32**,** 1777-1782.

Magrì, D., Limongelli, G., Re, F., Agostoni, P., Zachara, E., Correale, M., Mastromarino, V., Santolamazza, C., Casenghi, M., and Pacileo, G. (2016). Cardiopulmonary exercise test and sudden cardiac death risk in hypertrophic cardiomyopathy. *Heart***,** heartjnl-2015-308453.

Martin, D., and O'kroy, J. (1993). Effects of acute hypoxia on the VO2 max of trained and untrained subjects. *J. Sports Sci.* 11**,** 37-42.

Mourot, L., Perrey, S., Tordi, N., and Rouillon, J.D. (2004). Evaluation of fitness level by the oxygen uptake efficiency slope after a short-term intermittent endurance training. *Int. J. Sports Med.* 25**,** 85-91.

Ozcelik, O., and Kelestimur, H. (2004). Effects of acute hypoxia on the estimation of lactate threshold from ventilatory gas exchange indices during an incremental exercise test. *Physiol. res.* 53**,** 653-659.

Romer, L.M., Mcconnell, A.K., and Jones, D.A. (2002a). Effects of inspiratory muscle training on time-trial performance in trained cyclists. *J. Sports Sci.* 20**,** 547-590.

Romer, L.M., Mcconnell, A.K., and Jones, D.A. (2002b). Inspiratory muscle fatigue in trained cyclists: effects of inspiratory muscle training. *Med. Sci. Sports Exerc.* 34**,** 785-792.

Roussos, C. (1985). Function and fatigue of respiratory muscles. *Chest* 88**,** 124S-132S.

Rusko, H., Tikkanen, H., and Peltonen, J. (2004). Altitude and endurance training. *J. Sports Sci.* 22**,** 928-945.

Salazar-Martínez, E., Terrados, N., Burtscher, M., Santalla, A., and Orellana, J.N. (2016). Ventilatory efficiency and breathing pattern in world-class cyclists: A three-year observational study. *Respir. Physiol. Neurobiol.* 229**,** 17-23.

Sheel, A.W. (2002). Respiratory muscle training in healthy individuals. *Sports Med.* 32**,** 567-581.

Sun, X.-G., Hansen, J.E., Garatachea, N., Storer, T.W., and Wasserman, K. (2002). Ventilatory efficiency during exercise in healthy subjects. *Am. J. Respir. Crit. Care Med.* 166**,** 1443-1448.

Townsend, N.E., Gore, C.J., Hahn, A.G., Mckenna, M.J., Aughey, R.J., Clark, S.A., Kinsman, T., Hawley, J.A., and Chow, C.-M. (2002). Living high-training low increases hypoxic ventilatory response of well-trained endurance athletes. *J. Appl. Physiol.* 93**,** 1498-1505.

Volianitis, S., Mcconnell, A.K., Koutedakis, Y., Mcnaughton, L.R., Backx, K., and Jones, D.A. (2001). Inspiratory muscle training improves rowing performance. *Med. Sci. Sports Exerc.* 33: 803– 809.

Warner, M.M., and Mitchell, G.S. (1990). Ventilatory responses to hyperkalemia and exercise in normoxic and hypoxic goats. *Respir. Physiol.* 82**,** 239-249.

Wells, G.D., Plyley, M., Thomas, S., Goodman, L., and Duffin, J. (2005). Effects of concurrent inspiratory and expiratory muscle training on respiratory and exercise performance in competitive swimmers. *Eur. J. Appl. Physiol.* 94**,** 527-540.

Winkelmann, E.R., Chiappa, G.R., Lima, C.O.C., Viecili, P.R.N., Stein, R., and Ribeiro, J.P. (2009). Addition of inspiratory muscle training to aerobic training improves cardiorespiratory responses to exercise in patients with heart failure and inspiratory muscle weakness. *Am. Heart* 158**,** 768. e761-768. e767.

ESTUDIO III

INFLUENCE OF HIGH-INTENSITY INTERVAL TRAINING ON VENTILATORY EFFICIENCY IN TRAINED ATHLETES

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INFLUENCE OF HIGH-INTENSITY INTERVAL TRAINING ON VENTILATORY EFFICIENCY IN TRAINED ATHLETES

ABSTRACT

The aim of this study was to investigate the effects of 3-week high-intensity interval training (HIT) on ventilatory efficiency (V_E/VCO_2 slope) in athletes. Sixteen male well-trained (67.72 ml·kg·min^{-1}) sport students participated in this study. Each participant performed an incremental exercise test with gas analysis (i.e. V_E, VO_2) and a 400 m field test (T400m) before and after the 3-week intervention period. HIT group (HITG) performed 11 HIT sessions consisting of four 4-min interval bouts at an exercise intensity of 90–95% of the VO_{2max}, separated by 4-min active recovery periods (work/rest ratio = 1:1). No changes were found in the parameters studied. Ventilatory efficiency (up to VT_2 and up to exhaustion) did not show any change in HITG after training intervention (ES=0.24 HITG and ES=0.21 CG). No significant changes were observed in V_{Emax}, but HITG showed an increase in V_{Emax} after training (ES=0.38). VO_{2max} and T400m did not show a statistical improvement after the training period (no interaction time x group) (ES=0.43 and ES=0.75 respectively). These results do not support the hypothesis that 3-week of HIT could modify the ventilatory efficiency response in well-trained athletes. Furthermore, they show the lack of relationship between ventilatory efficiency and sport performance.

Key words: V_E/VCO_2 slope / HIT / ventilation / chemosensitivity / athletes

1. Introduction

High-intensity interval training (HIT) is defined as either repeated short (<45 s) to long (2-4 min) bouts of rather high-but not maximal intensity exercise, or short (<10 s, repeated-sprint sequences (RSS)) or long (>20-30 s, sprint interval session (SIT) all-out sprints interspersed with recovery periods (Buchheit & Laursen, 2013). HIT has increased its popularity during the last years as an effective method to improve exercise performance (Breil, Weber, Koller, Hoppeler, & Vogt, 2010). The mechanisms proposed which could explain the improvements in sport performance after HIT programs are: increments in VO_{2max} (Breil et al., 2010; Helgerud et al., 2007), changes in plasma volume (Richardson, Verstraete, Johnson, Luetkemeier, & Stray-Gundersen, 1996), changes in hormonal and metabolic response (Wahl, Mathes, Achtzehn, Bloch, & Mester, 2014) or increments in skeletal muscle oxidative capacity (Egan et al., 2010; Gibala et al., 2006). However, the influence of HIT on ventilatory parameters is still discussed controversially. Studies which included ventilatory variables in their analysis were only focused on ventilation (V_E). V_E determined at a fixed submaximal speed did not change after 12-week of HIT in athletes who completed the training intervention (Kilen et al., 2014). Maximum ventilation (V_{Emax}) and maximal breathing frequency (f_{Rmax}) did not change after 3-week of HIT neither in normoxia or hypoxia in basketball players (Czuba et al., 2013). Specifically, the influence of HIT on ventilatory efficiency has been only studied in patients without any changes on this variable after the training interventions (Cardozo, Oliveira, & Farinatti, 2015; Myers et al., 2012). The limited number of available studies in this area has reported that ventilatory efficiency generally improves after training, although this is not an entirely consistent finding [11]. In athletes, no changes on ventilatory efficiency (measured as V_E/VCO_2 slope) were found after 16-week of no controlled training in juvenile cyclists (Brown, Raman, Schlader, & Stannard, 2013) and after 3 competitive seasons in world-class cyclists (Salazar-Martínez, Terrados, Burtscher, Santalla, & Orellana, 2016). In order to better clarify the influence of exercise on ventilatory efficiency in athletes, the present pilot study aimed to investigate the effects of 3-week of HIT on V_E/VCO_2 slope in athletes. We hypothesized that 3-week of HIT could promote changes in the ventilatory efficiency response of well-trained athletes due to the inter-individual adjustment in breathing pattern described at high exercise intensities (Gravier, Delliaux, Delpierre, Guieu, & Jammes, 2013) and the high ventilatory requirements developed during HIT sessions.

2. Methods

2.1. Participants

Sixteen male well-trained sport students participated in the study (Table I). Possible exclusion criteria were all types of acute and chronic diseases or smoking. The study was carried out according to the Declaration of Helsinki and was approved by the Institutional Review Board of the Department of Sport Science (University Innsbruck). All participants gave written informed consent to participate in the study. Some of these participants were included previously in the sample size of other study (Menz, Strobl, Faulhaber, Gatterer, & Burtscher, 2015). However, we carried out a new analysis about ventilatory efficiency and performance variables never published before.

Table 1: Baseline age and physical characteristics of the training and the control group.

	HIT (n=8)	CG (n=8)
Age (years)	25.6±3.2	25±3.4
Height (cm)	181.6±5.8	178.7±4.9
Weight (kg)	74.2±5.5	75.2±6.3
VO_{2max} (ml·kg·min^{-1})	68.4±2.7	67±6.5

Data are presented as Mean ± SD

2.2. Design

The study was designed as a randomized controlled training study including HIT and control group (HITG and CG, respectively) and two measurement times (pre-training *vs* post-training). Baseline measurements included a laboratory incremental treadmill test and a 400m field test. After baseline measurements, the participants were randomly assigned, stratified by VO_{2max} either to the HITG or the CG. The HITG started the 3-week HIT program whereas CG maintained their usual training during this period. In particular, they were advised not to include additional high-intensity training. The training data for the HITG and the CG were recorded in a training log book and the total endurance training loads were determined according to (Foster et al., 2001) as perceived exertion × endurance training session time (Table II).

2.3. Methodology

2.3.1. Treadmill and 400m testing

The treadmill protocol was performed according to Burtscher, Gatterer, Faulhaber, Gerstgrasser, and Schenk (2010). Gas analysis was performed using an open spirometric system (Oxycon Mobile, Care Fusion, Würzburg, Germany) which was calibrated before each measurement. Cardio-respiratory parameters (i.e. V_E, VO_2, VCO_2, HR) were recorded breath by breath during the ergospirometry. A test was considered maximal when three of the four criteria proposed by (Cunha, Midgley, Monteiro, & Farinatti, 2010) were fulfilled. Additionally, participants carried out a 400m filed test. The time to complete the 400m (T400m) was selected as a performance variable. Athletes were encouraged to achieve their best performance.

2.3.2. HIT program

The HITG performed 11 HIT sessions during the 3-week HIT period. Each HIT session consisted of four 4-min interval bouts at an exercise intensity of 90–95% of the VO_{2max}, separated by 4-min active recovery periods (work/rest ratio = 1:1). In the first week, athletes completed 3 HIT sessions and in the following 2 weeks four HIT sessions each week. Training intensity was controlled by continuous heart rate (HR) monitoring (Polar, Kempele, Finland) and was equivalent to the HR at 90-95% of their VO_{2max} (García-Pallarés J.; Morán-Navarro, 2012). The rating of perceived exertion (RPE) was determined according to the Borg scale (6–20) (Borg, 1982).

Table 2: Training data (all endurance training performed during the 3-week training period) for HITG and CG.

	HIT group (n=8)			CG (n=8)		
	Endurance	Borg	Trimp	Endurance	Borg	Trimp
Week 1	329.1±134.5	13.7±1.9	3570.1±1914.8	331.2±95.8	13.2±0.9	4431.8±1549.3
Week 2	248±90.4	14.1±1.7	3163.5±1320.6	272.5±196.2	14.1±2.8	3522.2±2718.2
Week 3	297.5±123.2	14.5±1.3	4062.1±1632.3	390.7±192.7	14.8±4.1	5129.7±2439.8
Total	874.6±212.8	14.1±1.1	13,098.8±4734.5	994.5±418.1	14±2.3	12,326.7±5706

Data are presented as Mean ± SD

TRIMP training impulse (perceived exertion × endurance training session time)

2.3.3. Ventilatory efficiency

The V_E/VCO_2 slope was calculated from the slope of the relationship between VCO_2 and V_E during each incremental exercise test. V_E/VCO_2 slope was calculated from the beginning of the test until the second ventilatory threshold (VT_2) and up to exhaustion. VT_2 was identified by an increase in the ventilatory equivalent of CO_2 ($EqCO_2$) and a decrease in end tidal partial pressure of carbon dioxide ($PETCO_2$) (Lucía, Hoyos, Pérez, & Chicharro, 2000).

2.4. Statistical analysis

Data are expressed as mean ± SD and with Cohen's d effect size (ES) for each variable. Normal distribution of data was tested by the Kolmogorov–Smirnov test. A two-way analysis (group × time) of variance (ANOVA) with repeated measurements was used to verify between-group changes. In addition, paired student's t tests were carried out to evaluate within-group effects. The relationships between variables were assessed by regression analyses. The level of significance was set at $P<0.05$ for each statistical analysis. An ES of <0.2 was considered small, 0.5 medium and >0.8 large.

3. Results

No changes were found in the parameters studied (Table III). Ventilatory efficiency (up to VT_2 and up to exhaustion) did not show any change in HITG after training intervention (ES=0.24 HITG and ES=0.21 CG). No significant changes were observed in V_{Emax} (ES=0.38). VO_{2max} and T400m did not show a statistical improvement after the training period (no interaction time x group) (ES=0.43 and ES=0.75 in HITG respectively).

Table 3: Ventilatory and performance variables analyzed during the intervention period in both groups.

	HIT group (n=8)		Effect Size (Cohen´s d)	CG (n=8)		Effect Size (Cohen´s d)	ANOVA (interaction) time x group
	Pre	Post		Pre	Post		
V_E/VCO_2 slope (up to VT_2)	28.1±2.2	27.4±2.3	0.24	26.1±3.2	26.5±3.1	0.21	0.522
V_E/VCO_2 slope (up to exhaustion)	30.8±2.7	30.5±3.3	0.11	30.2±3.4	30.4±3.8	0.07	0.757
V_{Emax} (l·min^{-1})	181.8±17.8	184±19.4	0.38	181.7±14.5	181.5±17.5	0.02	0.607
VO_{2max} (ml·kg·min^{-1})	68.4±2.7	69.8±1.8	0.43	67.1±6.5	66.8±5.7	0.05	0.246
T400m (seg)	59.6±1.6	58.3±1.9	0.75	60.8±4.6	60.2±3.9	0.25	0.557

V_E/VCO_2 slope, ventilatory efficiency; V_{Emax}, maximum ventilation; VO_{2max}, maximum oxygen uptake; T400m, time in 400m field test

* p<0.05

<0.2 small, 0.5 medium and >0.8 large effect size (ES)

4. Discussion

The main finding of this study was that 3-week of HIT did not modify the ventilatory efficiency response of well-trained athletes. This is the first study to report reference of HIT on ventilatory efficiency in well-trained athletes. These results also suggest the lack of relationship between ventilatory efficiency and sport performance. This is an important finding, as it might indicate that ventilatory efficiency could be an inborn characteristic which response relative stable in healthy subjects independently of the improvements reported in sport performance. These results agree with recent evidence reported in athletes (Brown et al., 2013; Salazar-Martínez, Gatterer, Burtscher, Naranjo Orellana, & Santalla, 2017; Salazar-Martínez et al., 2016).

Conditions where the CO_2 production is elevated, as high intensity exercise, seem to play an essential role in the ventilatory control (Milsom, Abe, Andradeb, & Tattersall, 2004). Regular exercise training has been suggested as an effective stimulus which could modify CO_2 chemosensitivity in athletes (Kelley, Laufe, Millman, & Peterson, 1984; Miyamura & Ishida, 1990; Ohkuwa, Fujitsuka, Utsuno, & Miyamura, 1980). However, uncontrolled training did not have any effect on ventilatory efficiency response in athletes (Brown et al., 2013; Salazar-Martínez et al., 2016). In patients, ventilatory efficiency did not change after HIT sessions (Cardozo et al., 2015; Myers et al., 2012). We measured ventilatory efficiency not only up to VT_2, but also up to exhaustion for investigating its response after the quimio-compensation point where the highest rates of CO_2 production are developed for investigating whether this stimulus has any effect on V_E*vs*VCO_2 relationship. However, HIT did not modify ventilatory efficiency response up to VT_2 neither up to exhaustion (Table III). In the same way, maximum VCO_2 production did not show a significant change after training period. These results and the evidence reported before could indicate that the efficiency of the CO_2 elimination during exercise might move within very tight ranges, could be set by the system and might be hardly modified through training (Salazar-Martínez et al., 2016). According to that, increments in CO_2 production are linked to proportional increment in V_E in spite of a possible inter-individual adjustment in breathing patter at high exercise intensities (Gravier et al., 2013). This relationship is hard to modify with training, even with training sessions developed at high CO_2 production ratios (~5.5 l·min-^{1}) and with high ventilatory requirements (~185 l·min-^{1}). In addition to this finding, our results suggest the lack of relationship between ventilatory efficiency and sport performance (Figure 1, 2). Our results agree with previous evidence reported (Brown et al., 2013; Salazar-Martínez et al., 2016), and indicate that the ability to eliminate CO_2 during exercise (greater ventilatory efficiency) does not influence the capacity for achieving a high sport performance (Figure 1, 2).

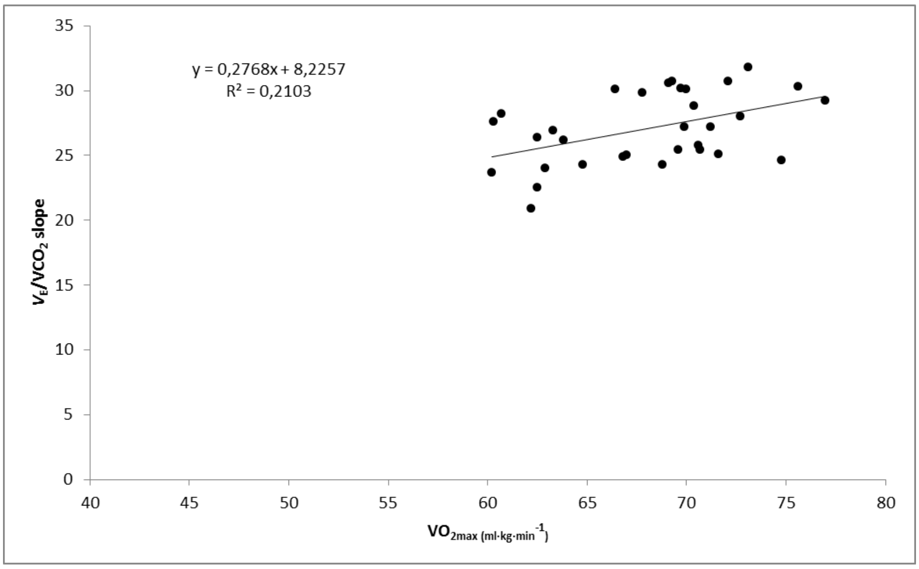

Figure 1: Relationship between ventilatory efficiency (V_E/VCO_2 slope) and maximum oxygen uptake (VO_{2max}) with data from both groups and both test.

Although the mechanisms which could explain the sport performance improvements after HIT have been already studied before, from our knowledge this is the first time that it has been studied the influence of HIT on ventilatory efficiency in well-trained athletes. With reference to V_{Emax}, VO_{2max} and T400m, HIT could have had a positive effect on these variables (medium-large ES, Table III). However, further studies are necessary in order to better clarify this point.

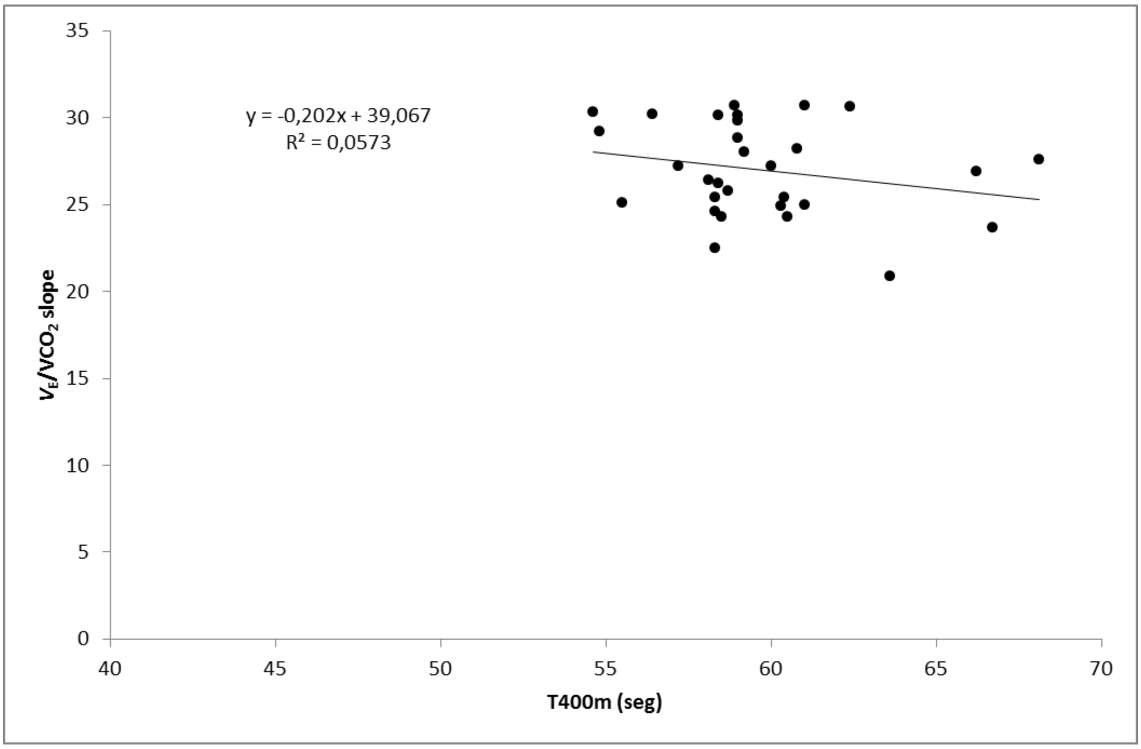

Figure 2: Relationship between ventilatory efficiency (V_E/VCO_2 slope) and the time in 400m test (T400m) with data from both groups and both test.

Few limitations must be addressed. Firstly, it could be possible that the intervention period (3-weeks) had not been enough to promote changes on ventilatory response. Lastly, the small sample size could have not been large enough to detect changes in sport performance variables.

5. Conclusions

Present preliminary data suggest that high-intensity interval training might be beneficial for improve performance in only 3-weeks. This finding may have relevance for athletes planning to complete a high intensity in a short time period. However, these results do not support the hypothesis that 3-week of HIT could modify the ventilatory efficiency of well-trained athletes. Furthermore, they show a lack of relationship between ventilatory efficiency and sport performance. Thus, this pilot study strengthen the previous evidence reported which support the idea that ventilatory efficiency could be an inborn characteristic which react in a tight ranges independently of sport performance improvements.

REFERENCES

Borg, G. A. (1982). Psychophysical bases of perceived exertion. *Med sci sports exerc, 14*(5), 377-381

Breil, F. A., Weber, S. N., Koller, S., Hoppeler, H., & Vogt, M. (2010). Block training periodization in alpine skiing: effects of 11-day HIT on VO2max and performance. *European journal of applied physiology, 109*(6), 1077-1086

Brown, S. J., Raman, A., Schlader, Z., & Stannard, S. R. (2013). Ventilatory efficiency in juvenile elite cyclists. *Journal of Science and Medicine in Sport, 16*(3), 266-270

Buchheit, M., & Laursen, P. B. (2013). High-intensity interval training, solutions to the programming puzzle. *Sports medicine, 43*(5), 313-338

Burtscher, M., Gatterer, H., Faulhaber, M., Gerstgrasser, W., & Schenk, K. (2010). Effects of intermittent hypoxia on running economy. *International journal of sports medicine, 31*(09), 644-650

Cardozo, G. G., Oliveira, R. B., & Farinatti, P. T. (2015). Effects of high intensity interval versus moderate continuous training on markers of ventilatory and cardiac efficiency in coronary heart disease patients. *The Scientific World Journal, 2015*

Cunha, F., Midgley, A., Monteiro, W., & Farinatti, P. (2010). Influence of cardiopulmonary exercise testing protocol and resting VO2 assessment on% HRmax,% HRR,% VO2max and% VO2R relationships. *International journal of sports medicine, 31*(05), 319-326

Czuba, M., Zając, A., Maszczyk, A., Roczniok, R., Poprzęcki, S., Garbaciak, W., & Zając, T. (2013). The effects of high intensity interval training in normobaric hypoxia on aerobic capacity in basketball players. *Journal of human kinetics, 39*(1), 103-114

Egan, B., Carson, B. P., Garcia-Roves, P. M., Chibalin, A. V., Sarsfield, F. M., Barron, N., . . . O'Gorman, D. J. (2010). Exercise intensity-dependent regulation of peroxisome proliferator-activated receptor γ coactivator-1α mRNA abundance is associated with differential activation of upstream signalling kinases in human skeletal muscle. *The Journal of physiology, 588*(10), 1779-1790

Foster, C., Florhaug, J. A., Franklin, J., Gottschall, L., Hrovatin, L. A., Parker, S., . . . Dodge, C. (2001). A new approach to monitoring exercise training. *The Journal of Strength & Conditioning Research, 15*(1), 109-115

García-Pallarés J.; Morán-Navarro, R. (2012). Propuesta metodológica para el entrenamiento de la resistencia cardiorrespiratoria. *Journal of Sport and Health Research, 4*(2), 119-136

Gibala, M. J., Little, J. P., Van Essen, M., Wilkin, G. P., Burgomaster, K. A., Safdar, A., . . . Tarnopolsky, M. A. (2006). Short-term sprint interval versus traditional endurance training: similar initial adaptations in human skeletal muscle and exercise performance. *The Journal of physiology, 575*(3), 901-911

Gravier, G., Delliaux, S., Delpierre, S., Guieu, R., & Jammes, Y. (2013). Inter-individual differences in breathing pattern at high levels of incremental cycling exercise in healthy subjects. *Respiratory physiology & neurobiology, 189*(1), 59-66

Helgerud, J., Hoydal, K., Wang, E., Karlsen, T., Berg, P., Bjerkaas, M., . . . Bach, R. (2007). Aerobic High-Intensity Intervals Improve VO~ 2~ m~ a~ x More Than Moderate Training. *Medicine and science in sports and exercise, 39*(4), 665

Kelley, M. A., Laufe, M. D., Millman, R. P., & Peterson, D. D. (1984). Ventilatory response to hypercapnia before and after athletic training. *Respiration physiology, 55*(3), 393-400

Kilen, A., Larsson, T. H., Jørgensen, M., Johansen, L., Jørgensen, S., & Nordsborg, N. B. (2014). Effects of 12 weeks high-intensity & reduced-volume training in elite athletes. *PloS one, 9*(4), e95025

Lucía, A., Hoyos, J., Pérez, M., & Chicharro, J. L. (2000). Heart rate and performance parameters in elite cyclists: a longitudinal study. *Medicine and Science in Sports and Exercise, 32*(10), 1777-1782

Menz, V., Strobl, J., Faulhaber, M., Gatterer, H., & Burtscher, M. (2015). Effect of 3-week high-intensity interval training on VO2max, total haemoglobin mass, plasma and blood volume in well-trained athletes. *European journal of applied physiology, 115*(11), 2349-2356

Milsom, W. K., Abe, A. S., Andradeb, D. V., & Tattersall, G. J. (2004). Evolutionary trends in airway CO 2/H+ chemoreception. *Respiratory physiology & neurobiology, 144*(2), 191-202

Miyamura, M., & Ishida, K. (1990). Adaptive changes in hypercapnic ventilatory response during training and detraining. *European journal of applied physiology and occupational physiology, 60*(5), 353-359

Myers, J., Gademan, M., Brunner, K., Kottman, W., Boesch, C., & Dubach, P. (2012). Effects of high-intensity training on indices of ventilatory efficiency in chronic heart failure. *Journal of cardiopulmonary rehabilitation and prevention, 32*(1), 9-16

Ohkuwa, T., Fujitsuka, N., Utsuno, T., & Miyamura, M. (1980). Ventilatory response to hypercapnia in sprint and long-distance swimmers. *European journal of applied physiology and occupational physiology, 43*(3), 235-241

Richardson, R. S., Verstraete, D., Johnson, S., Luetkemeier, M., & Stray-Gundersen, J. (1996). Evidence of a secondary hypervolemia in trained man following acute high intensity exercise. *International journal of sports medicine, 17*(04), 243-247

Salazar-Martínez, E., Gatterer, H., Burtscher, M., Naranjo Orellana, J., & Santalla, A. (2017). Influence of inspiratory muscle training on ventilatory efficiency and cycling performance in normorxia and hypoxia. *Frontiers in Physiology, 8*, 133

Salazar-Martínez, E., Terrados, N., Burtscher, M., Santalla, A., & Orellana, J. N. (2016). Ventilatory efficiency and breathing pattern in world-class cyclists: A three-year observational study. *Respiratory physiology & neurobiology, 229*, 17-23

Wahl, P., Mathes, S., Achtzehn, S., Bloch, W., & Mester, J. (2014). Active vs. passive recovery during high-intensity training influences hormonal response. *International journal of sports medicine, 35*(07), 583-589

ESTUDIO IV

VENTILATORY EFFICIENCY RESPONSE IS UNAFFECTED BY FITNESS LEVEL, ERGOMETER SETTINGS, AGE OR BODY MASS INDEX IN MALE-ATHLETES

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VENTILATORY EFFICIENCY RESPONSE IS UNAFFECTED BY FITNESS LEVEL, ERGOMETER SETTINGS, AGE OR BODY MASS INDEX IN MALE-ATHLETES

ABSTRACT

The aim of this study was to evaluate the ventilatory efficiency (V_E/VCO_2 slope) and the respiratory control (Vt/Ti slope) in a wide range of athletes and describe the influence of fitness level, age, ergometer or BMI in these parameters. Ninety-one males (30.4±10.53 years; 175.52±7.45 cm; 71.99±9.35 kg) were analysed retrospectively for the study. Ventilatory efficiency reacted similarly in athletes independently of the fitness level, the age, the BMI or the ergometer used for testing. No significant differences were found in V_E/VCO_2 slope and the Vt/Ti slope between variables analyzed ($P>0.05$). The slope of the predictive equations was similar in all cases studied in V_E/VCO_2 slope and the Vt/Ti slope. Moreover, the central control impulse of respiration was neither affected by the variables studied. These observations suggest that ventilatory efficiency (V_E/VCO_2 slope) could be a variable fixed by the respiratory system which tends to response similarly in athletes.

Key words: V_E/VCO_2 slope / Respiratory / Exercise Testing / Body composition / Efficiency

1. Introduction

Ventilatory efficiency can be defined as the relationship between carbon dioxide production (VCO_2) and ventilation (V_E) during an incremental exercise test (Arena, Myers, et al., 2007). It has been reported several ways for measuring ventilator efficiency (Brown, Raman, Schlader, & Stannard, 2013; Sun, Hansen, Garatachea, Storer, & Wasserman, 2002). However, using the slope of the relationship between VCO_2 and V_E (V_E/ VCO_2 slope) has been suggested as the best way for achieving a correct evaluation of the ventilatory efficiency during an incremental exercise test (Habedank et al., 1998). It adds information about the global ventilatory efficiency throughout entire test and not only at one metabolic rate as it happens with the equivalent of CO_2 (V_E/VCO_2) (Salazar-Martínez, Terrados, Burtscher, Santalla, & Naranjo Orellana, 2016).

Ventilatory efficiency has been widely studied in patients suffering congestive heart failure (CHF) or cardio-respiratory weakness (Arena, R., Myers, & Guazzi, 2008; Ingle et al., 2007; Laveneziana et al., 2010; Magrì et al., 2016). Values exceeding 34 are considered abnormal (Arena, Guazzi, & Myers, 2007; Arena, Myers, et al., 2007) or indicative of the inefficiency of the respiratory system (Brown et al., 2013). In healthy subjects, it has been reported an inter-variability in the values of the V_E/VCO_2 slope (from 19 to 32) (Sun et al., 2002).

The role and importance of ventilatory efficiency in human sport performance remains controversial. The matching of ventilation and perfusion in the lungs is the primary determinant of ventilatory efficiency (REF-101 VOL). Conditions where the CO_2 production is elevated, as exercise, seem to play an essential role in the ventilatory control (Milsom, Abe, Andradeb, & Tattersall, 2004). In this regard, it could be possible that a greater efficiency of CO_2 elimination during exercise might allow a higher sport performance. However, in elite-juvenile cyclists, no relationship has been found between maximal oxygen uptake (VO_{2max}) and V_E/VCO_2 slope (Brown et al., 2013). In the same way, it has been reported that changes in sport performance in world class-cyclists over three competitive seasons are not related to changes in V_E/VCO_2 slope (Salazar-Martínez et al., 2016). In syinchornonized swimmers, ventilatory efficiency kept unalterable by working conditions during the apneic episodes (Naranjo, Centeno, Carranza, & Cayetano, 2006). Data from our research group revealed that submaximal cycling performance was not related to ventilatory efficiency response (Salazar-Martínez, Gatterer, Burtscher, Naranjo Orellana, & Santalla, 2017). We hypothesized that increments in CO_2 production are linked to proportional increment in ventilation in spite of a fitness level.

Physiologic dead space (V_D/V_T) has been suggested as a variable that could modify ventilatory efficiency response (Sun et al., 2002). Age and anthropometric characteristics might influence V_D/V_T (Mummery et al., 2003). However, in children, ventilatory efficiency was not affected by sex despite differences in anthropometric characteristics (Guerrero, Naranjo, & Carranza, 2008). Same results were found in adults, no age or sex differences were found on ventilatory efficiency in healthy participants (Sun et al., 2002). However, from our knowledge there are not studies which evaluate ventilatory efficiency in athletes with different characteristics. Thus, measuring the influence of age and BMI on ventilatory efficiency is necessary in order to better clarify if there are differences between athletes with different characteristics.

Regarding type of ergometer, a test dependency has been reported in healthy woman, but not in males (Davis, Tyminski, et al., 2006). The authors explained these results due to a small amount of arterial hypoxemia coupled with a small amount of arterial hypercapnia in woman (Davis, Tyminski, et al., 2006). However, from our knowledge this is the only study mainly focused in this analysis. Thus, further evaluation in athletes is necessary in order to evaluate the influence of type of ergometer on ventilatory efficiency response.

Although, ventilatory efficiency has been already studied in healthy people, this variable has not been wide studied in athletes. Contrary to ventilatory efficiency, breathing pattern has been wide studied in athletes (Lucia, Carvajal, Calderon, Alfonso, & Chicharro, 1999; Lucia, Hoyos, Pardo, & Chicharro, 2001; Scheuermann & Kowalchuk, 1999). V_E can be decomposed into the product of two components: (a) central inspiratory activity, known as "driving" and expressed as the relationship between Vt and inspiratory time (Vt/Ti) and (b) the inspiration-expiration alternation, known as "timing", and expressed by the relationship between Ti and the total duration of the breathing cycle (Ti/Ttot) (Milic-Emili, 1982; Milic-Emili & Grunstein, 1976). Vt/Ti and Ti/Ttot responses during incremental exercise appear to be stable and independent of fitness level (Lucia et al., 1999; Naranjo et al., 2006). Studying the relationship between V_E, Vt/Ti and VCO_2 we could know if the central control of respiration makes ventilatory efficiency (V_E/VCO_2 slope) behaves similar in athletes independently of their characteristics.

Thus, the aim of this study was to evaluate ventilatory efficiency and respiratory control in a wide range of athletes and describe the influence of fitness level, age, ergometer or BMI in these parameters. In this regard, we hypothesize that ventilatory efficiency could be an inborn characteristic which responses similar in athletes independently of fitness level, age, ergometer or BMI.

2. Materials and methods

2.1. Subjects

From a large amount of incremental exercise tests carried out in our laboratory, we selected those which were carried out by healthy sporty people from different endurance sport disciplines (running, cycling, triathlon) and fitness level (amateur, semi-professional). Ninety-one active and healthy males (30.4±10.53 years; 175.52±7.45 cm; 71.99±9.35 kg) were analysed retrospectively for the study. Participants were classified in different groups depending on ergometer used for testing, BMI, age and VO_{2max} (Treadmill (n=37); Cycle ergometer (n=54); BMI: 18-25 (n=70); 25-30 (n=21); Age: 16-25 (n=40); 25-35 (n=16); 35-45 (n=23); >45 (n=12); VO_{2max}: <45VO_{2max} (37.8±7.4 $ml·kg^{-1}·min^{-1}$; n=43); >45 VO_{2max} 51.9±5.1 $ml·kg^{-1}·min^{-1}$; (n=48)). Fitness level classification was set according Paap and Takken (2014) proposal. Cardio-respiratory variables are shown in Table 1.

Participants were tested in our laboratory for different previous proposes. All previous studies were approved by the ethical committee of Pablo Olavide University and conformed to standards of treatment of human participants in research as outlined in the Fifth Declaration of Helsinki. Participants were informed (both in writing and orally) about all testing and training procedures and gave their written informed consent to participate prior to entering the study.

Table 1: Maximum cardio-respiratory values during the incremental exercise test (n=91)

	VO_2 (ml·min^{-1})	VCO_2 (ml·min^{-1})	f_R (br·min^{-1})	VT (ml)	V_E (l·min^{-1})	Ti/Ttot	Vt/Ti (ml·sec^{-1})	$PETCO_2$ (mmHg)
Mean	3219.8	4051.9	51.2	2240.4	112.8	0.41	4823.1	43.6
SD	571.1	808.2	11.6	424.6	26.2	0.05	962.6	6.6

SD, standard deviation; VO_2, oxygen uptake; VCO_2, carbon dioxide output; f_R, breathing frequency; Vt, tidal volume; V_E, ventilation; Ti/Ttot, timing; Vt/Ti, driving; $PETCO_2$, end tidal pressure of carbon dioxide.

2.2. Procedures

From the tests carried out in our laboratory we selected those performed with the same protocol on cycle ergometer (Ergoselek 200, Ergoline, Germany) or on treadmill (Ergorun 8, Down electronics, Germany). Each participant performed a maximum incremental exercise tests with gas analysis. During each test, oxygen uptake (VO_2), carbon dioxide output (VCO_2), respiratory exchange ratio (RER), ventilation (V_E), breathing frequency (f_R), tidal volume (VT), oxygen equivalent ($EqVO_2$), carbon dioxide equivalent ($EqCO_2$), driving (Vt/Ti) and timing (Ti/Ttot) were recorded every 5 seconds breath by breath with a gas analyser (Cpx última, medical graphics, USA). The system was calibrated prior to each test with gas mixtures of known concentration. After 4 min of warming up, participants started the test at 50W and then the load was increased by 25W each minute until volitional exhaustion on cycle ergometer. On treadmill, after 4 min of warming up the participants started the test at 7 km/h and the velocity was increased by 1 km/h each minute until volitional exhaustion. Tests were carried out under similar and controlled environmental conditions (20-25°C; 45-55% relative humidity). Achievement of maximal oxygen uptake (VO_{2max}) was accepted when a plateau was found in the relationship between VO_2 and power output or when three of the four criteria for maximal VO_{2max} were obtained (Howley, Bassett, & Welch, 1995).

2.3. Ventilatory efficiency and breathing pattern

The ventilatory efficiency of each subject was calculated from the slope of the relationship between VCO_2 and V_E during each test. To exclude the influence due to respiratory compensation for acidosis during highly intensive exercise, the V_E/VCO_2 slope was determined from the beginning of the test until the second ventilatory threshold (VT_2). VT_2 was identified using the criteria of increase in both ventilatory equivalents - EqO_2 and $EqCO_2$ - and end tidal partial pressure of oxygen ($PETO_2$) with no concomitant increase in end tidal partial pressure of carbon dioxide ($PETCO_2$) or decrease in $PETCO_2$ (Lucía, Hoyos, Pérez, & Chicharro, 2000; Skinner & McLellan, 1980). The value of the slope representing the relationship between V_E and Vt/Ti during each test (Vt/Ti slope) was used to test the central component of respiration.

2.4. Statistical analysis

Data are expressed as mean ± SD and with Cohen's d effect size (ES) for each variable. Subjects were included in different groups depend on fitness level, ergometer used for testing, BMI and age. The normal distribution of the data in each group was checked by means of the Shapiro–Wilk test. The homogeneity of variance was evaluated by Levene´s test. To compare the mean values obtained of V_E/VCO_2 slope and Vt/Ti slope in each group the following statistical tests were carried out. Student's T-Test for independent samples was used to compare fitness level groups and type of ergometer groups. Kruskal–Wallis H-test was carried out to compare mean values between BMI groups. One-way ANOVA test were used to compare mean values between age groups. The Bonferroni test was selected as a post hoc test. Linear regression analysis was performed for each group between V_E (dependent variable) and VCO_2 (independent variable) and Vt/Ti (dependent variable) with data from each subject. Effect sizes (ES) were also calculated using Cohen´s d. The level of significance was set at $P<0.05$ for each statistical analysis. An ES of $d<0.2$ was considered small, 0.5 medium and $d>0.8$ large (Cohen, 1988).

3. Results

Data on the ventilatory efficiency and ventilatory control evaluation are shown in Table 2. The statistical analysis found non-significant differences (P>0.05) both for the V_E/VCO_2 slope and Vt/Ti slope for all the variables included in the analysis (ergometer, BMI, age, and fitness level). Effect size analysis showed a low ES between cycle-ergometer and treadmill testing on V_E/VCO_2 slope and Vt/Ti slope (0.29 and 0.09 respectively). Regarding BMI, a low-medium ES was found between groups in V_E/VCO_2 slope and Vt/Ti slope (0.46 and 0.24 respectively). No age effect was found in V_E/VCO_2 slope and in Vt/Ti slope (0.16 and 0.15 respectively). Fitness level showed a low ES for diffrences between groups in V_E/VCO_2 slope (0.33) and Vt/Ti slope (0.33). Table 3 shows the predicting equations for V_E/VCO_2 slope after regression and statistical analysis. The slope of the predictive equations was similar in all cases studied (Table 3). Figure 1 shows the regression lines for each variable studied.

Table 2: Comparison of Mean±SD values of the V_E/VCO_2 slope and Vt/Ti slope for the treadmill and cycleergometer cardiopulmonary exercise tests, the body mass index (BMI) ranges (18-25; 25-30), age ranges (16-25; 25-35; 35-45; >45) and fitness level (<45 VO_2max; >45 VO_2max) in athletes.

	ERGOMETER				**BMI ($kg \cdot m^{-2}$)**				**AGE (years)**						**FITNESS LEVEL: VO_2max ($ml \cdot kg^{-1} \cdot min^{-1}$)**			
	Cycle (n=37)	Treadmill (n=54)	p-value	Effect Size	18-25 (n=70)	25-30 (n=21)	p-value	Effect Size	16-25 (n=40)	25-35 (n=16)	35-45 (n=23)	>45 (n=12)	p-value	Effect Size	<45 VO_2max (n=43)	>45 VO_2max (n=48)	p-value	Effect Size
V_E/VCO_2 slope	23.6±3.8	24.8±4.4	0.146	0.29	24.5±4.1	22.6±4	0.067	0.46	24.3±3.8	22.9±4.5	24.1±4.6	25.6±3.7	0.146	0.16	23.4±4.2	24.8±4.1	0.111	0.33
Vt/Ti slope	38.7±6.5	39.4±6.3	0.592	0.09	38.8±6.3	40.4±7.1	0.336	0.26	38.8±6.4	38.4±6.2	40.7±6.6	38.1±6.7	0.416	0.15	40.5±6.3	38.3±6.3	0.100	0.33

*Significantly different between groups ($p < 0.05$).

§ Large effect size (ES≥0.8).

Table 3: Predictive equations for the ventilatory efficiency response.

	Predictive equations					
	a	b	r^2	r	Standard Error	p-value
Ergometer						
Cycle	25.81	0.964	0.929	0.964	0.07	0.000*
Treadmill	24.11	0.913	0.834	0.913	0.106	0.000*
BMI **(kg·m^{-2})**						
18-25	24.70	0.948	0.899	0.948	0.064	0.000*
25-30	24.66	0.950	0.903	0.950	0.125	0.000*
AGE **(years)**						
16-25 (n=48)	24.91	0.963	0.927	0.963	0.072	0.000*
25-35 (n=28)	24.49	0.936	0.875	0.936	0.137	0.000*
35-45 (n=23)	23.28	0.890	0.793	0.890	0.177	0.000*
>45 (n=12)	26.03	0.986	0.973	0.986	0.088	0.000*
FITNESS LEVEL: VO_{2max} (ml·kg^{-1}·min^{-1})						
<45 VO_{2max} (n=43)	24.12	0.945	0.893	0.945	0.094	0.000*
>45 VO_{2max} (n=62)	24.88	0.941	0.885	0.941	0.081	0.000*

* Level of significance ($p < 0.05$).

y = a · x + b (y=V_E (ventilation); x=VCO_2 (carbon dioxide output); a=V_E/VCO_2 slope; b= y-intercept)

4. Discussion

To the best of our knowledge, this is the first study which evaluates the influence of ergometer, age, BMI and fitness level on ventilatory efficiency in athletes. We hypothesized that ventilatory efficiency could behave independently of before mentioned variables in athletes. The main finding of this study was that ventilatory efficiency is not influenced by the ergometer used for testing, the athlete´s age, BMI or fitness level. These findings support the hypothesis that ventilatory efficiency could be an inborn characteristic which react independently of fitness level, anthropometric profile, age or the ergometer used for testing.

Ventilatory efficiency has been proposed as an effectiveness method to detect cardiorespiratory weakness and healthy problems (Arena, R. et al., 2008; Ingle et al., 2007; Magrì et al., 2016). Values exceeding 34 are indicative of the inefficiency of the cardiorespiratory system (Arena, Myers, et al., 2007; Chase et al., 2008). However, it is not as clear that athletes with better ventilatory efficiency are those who perform a high sport performance. In our study, no differences were found in V_E/VCO_2 slope between athletes with a low VO_{2max} *vs* high VO_{2max} (23.4±4.2 and 24.8±4.1, respectively). The slope of the predictive equations was also similar in both cases (24.12 and 24.88, respectively) (Table 3) (Figure 1). Similar mean values of efficiency were found in world-class cyclists over 3-year period (24.6±3.1; 23.6±2.7; 24.8±2.6) (Salazar-Martínez et al., 2016). Even though, these cyclists were tested with a totally different protocol (50W each 4 min), gas analyzer and they had a higher VO_{2max} (77.5±6.2 $ml \cdot kg^{-1} \cdot min^{-1}$), they showed similar values of ventilatory efficiency to our subjects. In this way, changes in sport performance (peak power output) were no related to changes in V_E/VCO_2 slope or VO_{2max} in world-class cyclists (Salazar-Martínez et al., 2016). In juvenile cyclists, no relationship was found between VO_{2max} and V_E/VCO_2 slope (Brown et al., 2013). No correlation was found between V_E/VCO_2 slope and VO_{2max} in sport students before and after inspiratory muscle training neither normoxia nor hypoxia (Salazar-Martínez et al., 2017). Thus, our results and the evidence reported before, help us to confirm the hypothesis that V_E/VCO_2 slope could not be a variable related to sport performance. In this regard, it has been suggested that if an athlete has poor cardio-respiratory efficiency (high V_E/VCO_2 slope) it has no bearing on their maximal ability to use oxygen (Brown et al., 2013) or achieving a high performance (Salazar-Martínez et al., 2016). Therefore, V_E/VCO_2 slope has not efficacy in quantifying the performance of the physiological systems which support an athlete's ability to perform at high oxygen uptakes (Brown et al., 2013).

In terms of age and BMI, controversial data about ventilatory efficiency has been reported. On the one hand, Sun et al. (2002) carried out an evaluation of ventilatory efficiency on healthy people without significant difference between sexes and ages. On the other hand, ventilatory efficiency showed a sex and age dependence in healthy subjects (Habedank et al., 1998). In children, ventilatory efficiency response was not affected by sex (Guerrero et al., 2008). In our study, we could not compare ventilatory efficiency between sexes due to the small sample size in females. Regarding age analysis, no differences were found between age groups in V_E/VCO_2 slope (Table 2). These results are in concordance with previous studies (Guerrero et al., 2008; Sun et al., 2002). Physiologic dead space (V_D/V_T) has been proposed as a variable that could modify ventilatory efficiency in healthy subjects (Sun et al., 2002). Maturation and age could modify the V_D/V_T (Mummery et al., 2003) and as a consequence ventilatory efficiency. In our subjects, the mean values obtained in age groups were similar to values measured in children (Guerrero et al., 2008) (Table 2). Thus, ventilatory efficiency might be a variable no affected by age or anthropometric characteristics in healthy athletes.

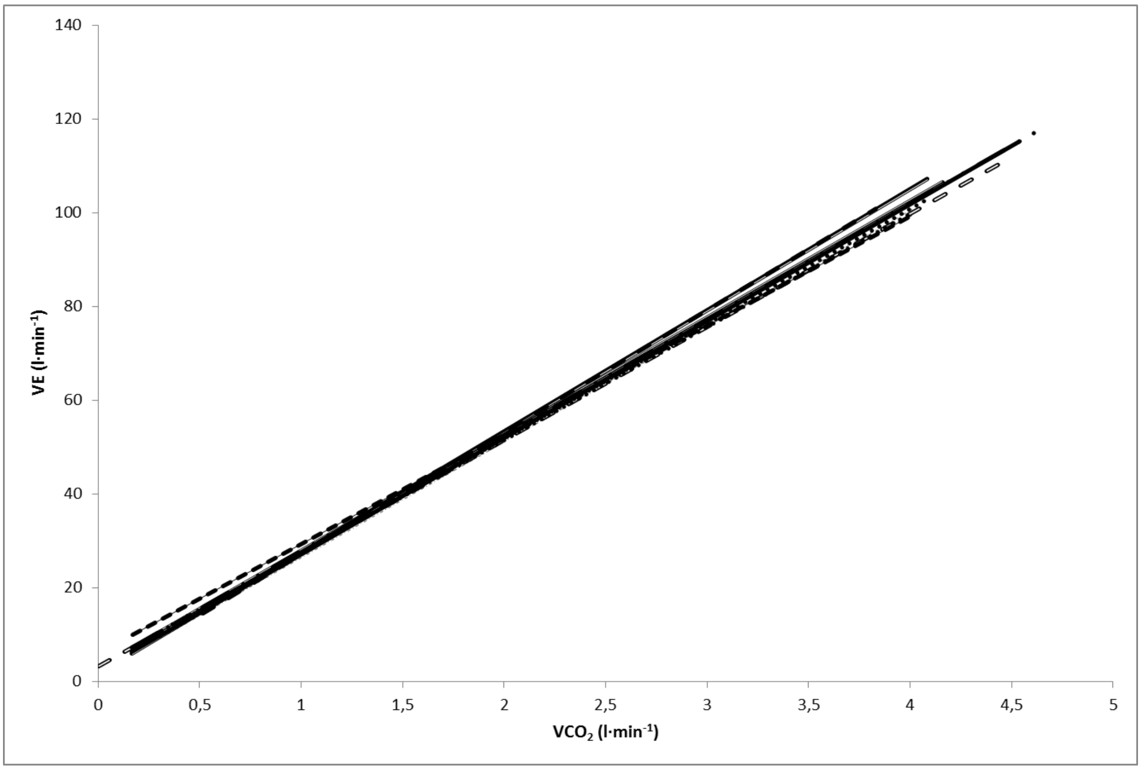

Figure 1: Evaluation of ventilatory efficiency (V_E/VCO_2 slope) showing regression lines measured in each group (Treadmill (n=37); Cycle ergometer (n=54); BMI: 18-25 (n=70); 25-30 (n=21); Age: 16-25 (n=40); 25-35 (n=16); 35-45 (n=23); >45 (n=12); VO_{2max}: <45VO_{2max} (n=43); >45 VO_{2max} (n=48)). All groups showed a similar linear adjustment.

With reference to type of ergometer, we did not find difference between subjects tested on treadmill or cycle ergometer on ventilatory efficiency response (Table 2). We compared world-class cyclist ventilatory efficiency data (Salazar-Martínez et al., 2016), who were tested with a different gas analyzer and with a different protocol (50W/4 min), with our subjects (25W/min). The mean values obtained were similar in both cases (~24). Same results were obtained in men but not in women, suggesting an independence of test mode evaluation (Davis, Tyminski, et al., 2006) and an independence of speed used in the test on ventilatory efficiency response (Davis, Sorrentino, Soriano, Pham, & Dorado, 2006). In the first study (Davis, Tyminski, et al., 2006), the protocol used (4 min of walking at 72 m·min^{-1} and 0% grade. At the end of minutes 4, 7 and 10, the speed was increased by 10 m·min^{-1}) was totally different to ours. In the second (Davis, Sorrentino, et al., 2006), they did not found different between the fast (25W/min) and the slow protocol (five work rate increments of equal size each 4 min). But one more time, the slope values reported (24.19 and 23.23 respectively) were in concordance with our results. In the same way, Sun et al. (2002) found an absence of the effect of laboratory site or ergometer in ventilatory efficiency evaluation, with a greater reproducibility for V_E/VCO_2 slope (online data supplement). The slope of the predictive equations was similar in all cases studied (Table 3). Accordingly to these results, type of ergometer or protocol used might not modify the ventilatory efficiency response in healthy athletes.

Furthermore ventilatory efficiency analysis, we carried out an analysis of driving component of respiration (Vt/Ti slope). As it occurs with V_E/VCO_2 slope, the increment in the driving impulse was similar in all our subjects and it was independent of age, fitness level, BMI or ergometer (Table 2). In all these cases, the increment in driving impulse was closely to ~40. This indicates that the increases in V_E during a progressive exercise are associated with a proportional increase in the inspiratory driving activity without any alteration in the relationship between inspiration and expiration, even at the highest working intensities (Figure 3) (Salazar-Martínez et al., 2016). Thus, the lineal relationship of V_E with Vt/Ti and VCO_2 suggest that the main factor conditioning the stability of ventilatory efficiency (as V_E/VCO_2 slope) could be the central impulse of respiration (Vt/Ti).

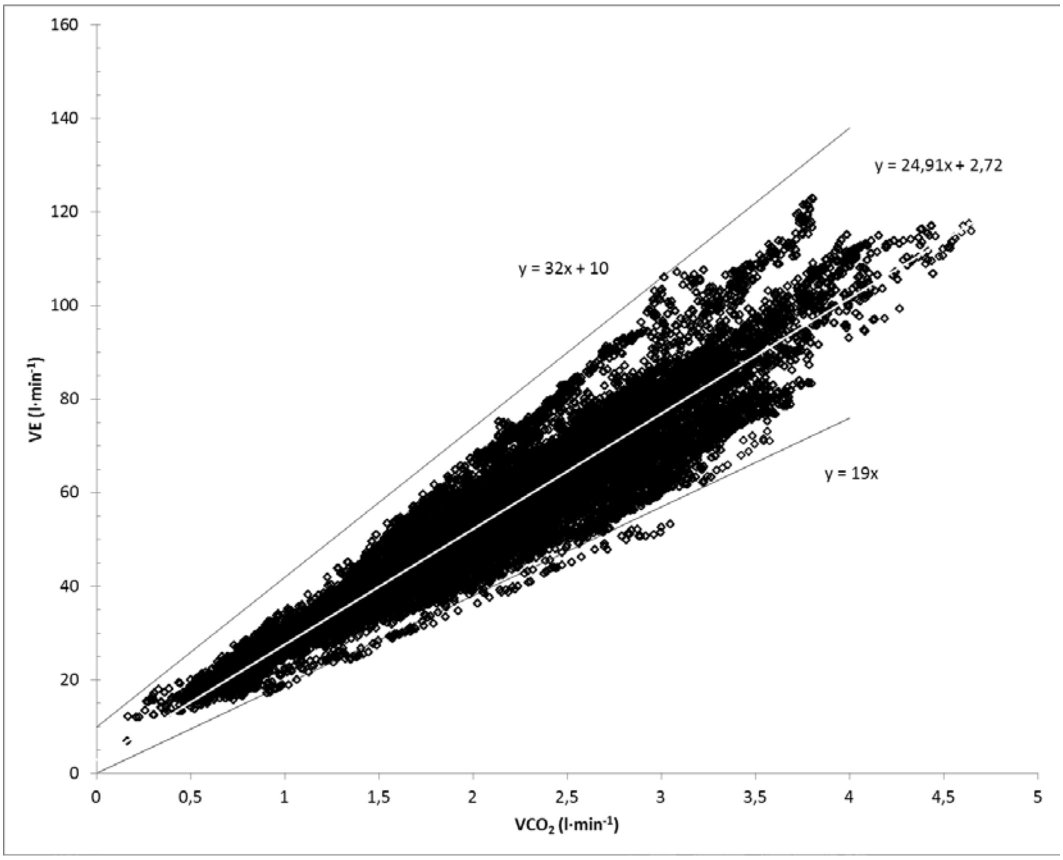

Figure 2: Graph showing the linear relation between carbon dioxide output (VCO_2) and ventilation (V_E) with data from all sample size (n=91). This can be used as a nomogram for assessing ventilatory efficiency in healthy athletes during exercise regardless of the ergometer, fitness level, age or body mass index.

Some limitations have to be addressed. First, this study was retrospectively and we could not measure body composition variables in our subjects. Further investigations taking into account body composition variables would be necessary in order to better clarify if body composition could influence ventilatory efficiency response. Lastly, we could not include females in our study due to the low sample size. New research to evaluate the influence of gender on ventilatory efficiency is necessary in order to better clarify the involvement of this variable on ventilatory efficiency response.

Based on the previous evidence reported and in our results, we propose a nomogram for assessing ventilatory efficiency (V_E/VCO_2 slope) (Figure 2). This nomogram might help to carry out a better evaluation of ventilatory efficiency in athletes completing the proposal of Naranjo et al. (2006). In addition, they could help to detect easily cardio-respiratory problems or deficiencies in respiration control when an incremental test is carried out in athletes.

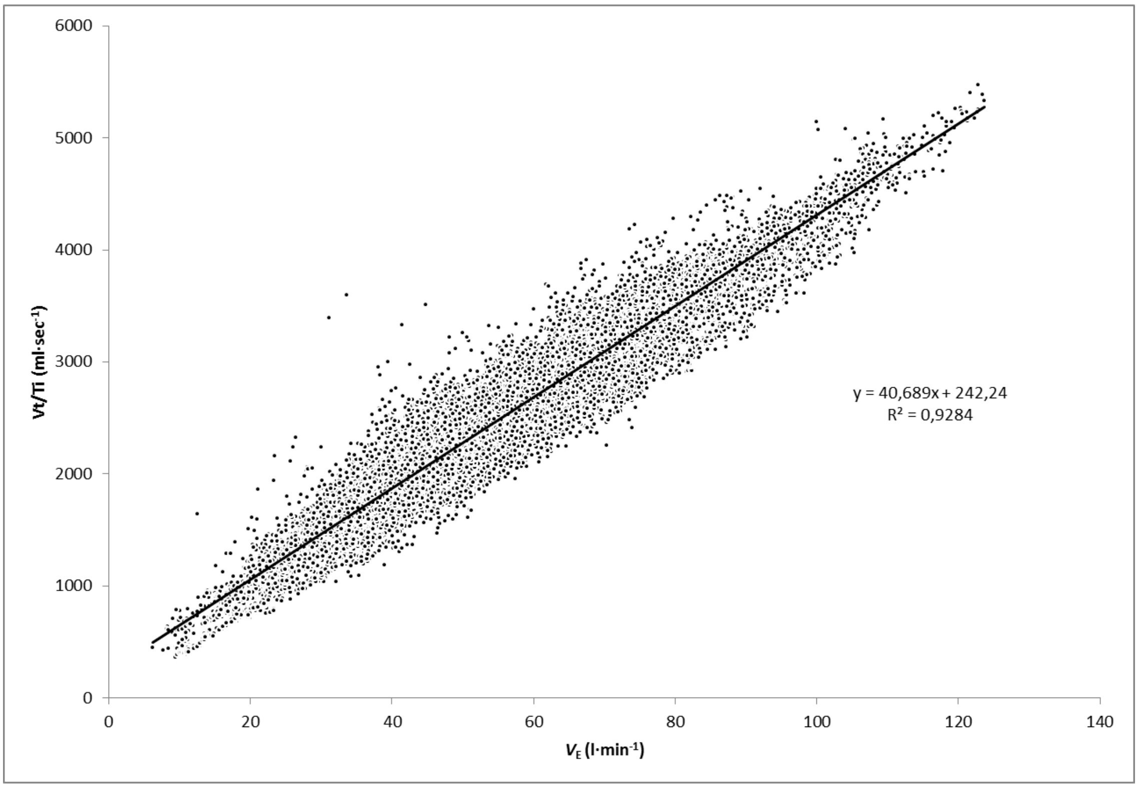

Figure 3: Graph showing the linear relation between and ventilation (V_E) and driving impulse (Vt/Ti) with data from all sample size (n=91). Central impulse of respiration responded similar in all participants regardless of the ergometer, fitness level, age or body mass index.

5. Conclusion

In summary, ventilatory efficiency reacted similarly in athletes independently of the fitness level, the age, the BMI or the ergometer used for testing. Moreover, the central control impulse of respiration was neither affected by the variables studied (Figure 3). These observations suggest that ventilatory efficiency (V_E/VCO_2 slope) could be a variable fixed by the respiratory system which tends to response similarly in athletes. Finally, ventilatory efficiency could be assessed easily during an incremental test in athletes using the nomogram proposed.

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Conflict of interest

No potential conflict of interest was reported by the authors.

REFERENCES

Arena, Guazzi, & Myers. (2007). Prognostic value of end-tidal carbon dioxide during exercise testing in heart failure. *Int J Cardiol, 117*(1), 103-108. doi: 10.1016/j.ijcard.2006.04.058

Arena, Myers, Hsu, Peberdy, Pinkstaff, Bensimhon, . . . Guazzi. (2007). The minute ventilation/carbon dioxide production slope is prognostically superior to the oxygen uptake efficiency slope. *J Card Fail, 13*(6), 462-469. doi: 10.1016/j.cardfail.2007.03.004

Arena, R., Myers, J., & Guazzi, M. (2008). The clinical and research applications of aerobic capacity and ventilatory efficiency in heart failure: an evidence-based review. *Heart Fail Rev, 13*(2), 245-269. doi: 10.1007/s10741-007-9067-5

Brown, S. J., Raman, A., Schlader, Z., & Stannard, S. R. (2013). Ventilatory efficiency in juvenile elite cyclists. *Journal of Science and Medicine in Sport, 16*(3), 266-270

Chase, P., Arena, R., Myers, J., Abella, J., Peberdy, M. A., Guazzi, M., & Bensimhon, D. (2008). Relation of the prognostic value of ventilatory efficiency to body mass index in patients with heart failure. *The American journal of cardiology, 101*(3), 348-352

Cohen, J. (1988). Statistical power analysis

Davis, J. A., Sorrentino, K. M., Soriano, A. C., Pham, P. H., & Dorado, S. (2006). Is ventilatory efficiency dependent on the speed of the exercise test protocol in healthy men and women? *Clinical physiology and functional imaging, 26*(2), 67-71

Davis, J. A., Tyminski, T. A., Soriano, A. C., Dorado, S., Costello, K. B., Sorrentino, K. M., & Pham, P. H. (2006). Exercise test mode dependency for ventilatory efficiency in women but not men. *Clinical physiology and functional imaging, 26*(2), 72-78

Guerrero, L., Naranjo, J., & Carranza, M. D. (2008). Influence of gender on ventilatory efficiency during exercise in young children. *J Sports Sci, 26*(13), 1455-1457. doi: 10.1080/02640410802255771

Habedank, D., Reindl, I., Vietzke, G., Bauer, U., Sperfeld, A., Gläser, S., . . . Kleber, F. X. (1998). Ventilatory efficiency and exercise tolerance in 101 healthy volunteers.

European journal of applied physiology and occupational physiology, 77(5), 421-426

Howley, E. T., Bassett, D. R., & Welch, H. G. (1995). Criteria for maximal oxygen uptake: review and commentary. *Medicine and science in sports and exercise, 27*(9), 1292-1301

Ingle, L., Goode, K., Carroll, S., Sloan, R., Boyes, C., Cleland, J. G., & Clark, A. L. (2007). Prognostic value of the VE/VCO2 slope calculated from different time intervals in patients with suspected heart failure. *Int J Cardiol, 118*(3), 350-355. doi: 10.1016/j.ijcard.2006.07.105

Laveneziana, P., Agostoni, P., Mignatti, A., Mushtaq, S., Colombo, P., Sims, D., . . . Jorde, U. P. (2010). Effect of Acute β-blocker Withholding on Ventilatory Efficiency in Patients With Advanced Chronic Heart Failure. *Journal of cardiac failure, 16*(7), 548-555

Lucia, A., Carvajal, A., Calderon, F. J., Alfonso, A., & Chicharro, J. L. (1999). Breathing pattern in highly competitive cyclists during incremental exercise. *Eur J Appl Physiol Occup Physiol, 79*(6), 512-521. doi: 10.1007/s004210050546

Lucia, A., Hoyos, J., Pardo, J., & Chicharro, J. L. (2001). Effects of endurance training on the breathing pattern of professional cyclists. *Jpn J Physiol, 51*(2), 133-141

Lucía, A., Hoyos, J., Pérez, M., & Chicharro, J. L. (2000). Heart rate and performance parameters in elite cyclists: a longitudinal study. *Medicine and Science in Sports and Exercise, 32*(10), 1777-1782

Magrì, D., Limongelli, G., Re, F., Agostoni, P., Zachara, E., Correale, M., . . . Pacileo, G. (2016). Cardiopulmonary exercise test and sudden cardiac death risk in hypertrophic cardiomyopathy. *Heart*, heartjnl-2015-308453

Milic-Emili, J. (1982). Recent advances in clinical assessment of control of breathing. *Lung, 160*(1), 1-17. doi: 10.1007/BF02719267

Milic-Emili, J., & Grunstein, M. M. (1976). Drive and timing components of ventilation. *Chest, 70*(1 Suppl), 131-133

Milsom, W. K., Abe, A. S., Andradeb, D. V., & Tattersall, G. J. (2004). Evolutionary trends in airway CO 2/H+ chemoreception. *Respiratory physiology & neurobiology, 144*(2), 191-202

Mummery, H. J., Stolp, B. W., deL Dear, G., Doar, P. O., Natoli, M. J., Boso, A. E., . . . Moon, R. E. (2003). Effects of age and exercise on physiological dead space during simulated dives at 2.8 ATA. *Journal of Applied Physiology, 94*(2), 507-517

Naranjo, J., Centeno, R. A., Carranza, M. D., & Cayetano, M. (2006). A test for evaluation of exercise with apneic episodes in synchronized swimming. *International journal of sports medicine, 27*(12), 1000-1004

Paap, D., & Takken, T. (2014). Reference values for cardiopulmonary exercise testing in healthy adults: a systematic review. *Expert review of cardiovascular therapy, 12*(12), 1439-1453

Salazar-Martínez, E., Gatterer, H., Burtscher, M., Naranjo Orellana, J., & Santalla, A. (2017). Influence of inspiratory muscle training on ventilatory efficiency and cycling performance in normorxia and hypoxia. *Frontiers in Physiology, 8*, 133

Salazar-Martínez, E., Terrados, N., Burtscher, M., Santalla, A., & Naranjo Orellana, J. (2016). Ventilatory efficiency and breathing pattern in world-class cyclists: A three-year observational study. *Respiratory Physiology & Neurobiology, 229*, 17-23

Scheuermann, B. W., & Kowalchuk, J. M. (1999). Breathing patterns during slow and fast ramp exercise in man. *Exp Physiol, 84*(1), 109-120

Skinner, J. S., & McLellan, T. H. (1980). The transition from aerobic to anaerobic metabolism. *Research quarterly for exercise and sport, 51*(1), 234-248

Sun, Hansen, Garatachea, Storer, & Wasserman. (2002). Ventilatory efficiency during exercise in healthy subjects. *American journal of respiratory and critical care medicine, 166*(11), 1443-1448

CAPÍTULO 7

CONCLUSIONES / CONCLUSIONS

SALAZAR-MARTÍNEZ E.

7- CONCLUSIONES [Conclusions]

7.1. CONCLUSIÓN GENERAL

Las principales conclusiones extraídas de todo el proceso de investigación llevado a cabo durante el desarrollo de esta Tesis Doctoral Internacional, son:

1. La eficiencia ventilatoria es un parámetro sujeto a poca variabilidad intra-sujeto.

2. Ni las grandes cargas de entrenamiento no controlado, ni protocolos de entrenamiento específicamente diseñados y controlados provocan cambios en la respuesta de la eficiencia ventilatoria en deportistas.

3. Independientemente de las características del sujeto evaluado y del instrumento usado para evaluar la eficiencia ventilatoria, ésta tiende a responder de forma similar en deportistas.

4. Aparentemente no existe una relación entre la respuesta de la eficiencia ventilatoria y el rendimiento físico y deportivo.

7.2. CONCLUSIONES ESPECÍFICAS RELACIONADAS CON LOS OBJETIVOS MARCADOS

1) No existe una relación entre la capacidad de eliminar CO_2 durante el ejercicio y la obtención de un mayor rendimiento deportivo (**estudios I, II, III, IV**).

2) La respuesta de la eficiencia ventilatoria está sujeta a la acción del centro respiratorio central el cual no permite una variabilidad inter e intra sujetos (**estudios I, IV**).

3) No existen diferencias significativas en la respuesta de la eficiencia ventilatoria entre sujetos de diferentes disciplinas deportivas, con diferentes niveles de entrenamiento, evaluados con diferentes ergómetros y con diferentes características antropométricas (**estudio IV**).

4) La respuesta de la eficiencia ventilatoria parece tender a mantenerse constante en deportistas sanos (**estudios I, II, III, IV**).

5) La respuesta de la eficiencia ventialtoria en condiciones de hipoxia (FiO_2=16.45) se ve modificada por la influencia de ésta en la ventilación (**estudio II**).

6) Un entrenamiento específico de los músculos inspiratorios no influye en sobre la respuesta de la eficiencia ventilatoria en condiciones normales de oxígeno (**estudio II**).

7) El entrenamiento específico de los músculos inspiratorios podría tener un efecto positivo sobre la respuesta de la eficiencia ventilatoria en hipoxia (**estudio II**).

8) El entrenamiento de la musculatura inspiratoria mejora el rendimiento en condiciones de hipoxia (**estudio II**).

9) Mejoras en el rendimiento deportivo tras un periodo de entrenamiento de los músculos inspiratorios, no están relacionadas con cambios en la eficiencia ventilatoria medida como VE/VCO_2 slope, pero sí medida a través de la OUES (**estudio II**).

10) La respuesta de la eficiencia ventilatoria durante el ejercicio se ve comprometida en situaciones ambientales de hipoxia (FiO_2=16.45) en comparación con las de normoxia (**estudio II**).

11) Un programa de tres semanas de entrenamiento interválico de alta intensidad no genera cambios en la respuesta de la eficiencia ventilatoria de sujetos entrenados (**estudio III**).

12) Las pequeñas mejoras detectadas en sujetos entrenados tras un programa de entrenamiento interválico de alta intensidad no están asociadas a cambios o mejoras en la eficiencia ventilatoria (**estudio III**).

HIPÓTESIS	OBJETIVOS	CONCLUSIONES
1- La eficiencia ventilatoria podría ser un factor determinante del rendimiento deportivo en sujetos sanos.	A. Conocer la relación entre la eficiencia ventilatoria y el rendimiento deportivo en deportistas (**estudios I, II, III, IV**).	1) No existe una relación entre la capacidad de eliminar CO_2 durante el ejercicio y la obtención de un mayor rendimiento deportivo **(estudios I, II, III, IV).**
	K. Conocer la respuesta de la eficiencia ventilatoria tras la aplicación de un programa de entrenamiento (**estudio II, III**).	11) Un programa de tres semanas de entrenamiento interválico de alta intensidad no genera cambios en la respuesta de la eficiencia ventilatoria de sujetos entrenados (**estudio II, III**).
		6) Un entrenamiento específico de los músculos inspiratorios no influye en sobre la respuesta de la eficiencia ventilatoria en condiciones normales de oxígeno (**estudio II**).
2- Cambios en el rendimiento deportivo podrían estar relacionados con cambios o mejoras en la eficiencia ventilatoria.	I. Evidenciar si mejoras en el rendimiento deportivo tras un periodo de entrenamiento de la musculatura respiratoria están relacionadas con mejoras en la eficiencia ventilatoria en deportistas (**estudio II**).	9) Mejoras en el rendimiento deportivo tras un periodo de entrenamiento de los músculos inspiratorios, no están relacionadas con cambios en la eficiencia ventilatoria medida como VE/VCO_2 slope, pero sí medida a través de la OUES (**estudio II**).
3- Cambios en la eficiencia ventilatoria podrían estar relacionados con cambios en el patrón respiratorio.	B. Comprender en mejor medida la influencia del centro respiratorio en la respuesta de la eficiencia ventilatoria durante el ejercicio (**estudios I, IV**).	2) La respuesta de la eficiencia ventilatoria está sujeta a la acción del centro respiratorio central el cual no permite una variabilidad inter e intra sujetos (**estudios I, IV**).
	D. Aportar una mayor evidencia científica sobre la respuesta de la eficiencia ventilatoria en ejercicio incremental en deportistas (**estudios I, II, III, IV**).	4) La respuesta de la eficiencia ventilatoria parece tender a mantenerse constante en deportistas sanos (**estudios I, II, III, IV**).

4- La eficiencia ventilatoria podría comportarse de manera similar en deportistas independientemente de las características de cada deportista.	C. Conocer si existen diferencias en la respuesta de la eficiencia ventilatoria en deportistas de diferentes disciplinas, características antropométricas, nivel de condición física y edad (**estudio IV**).	3) No existen diferencias significativas en la respuesta de la eficiencia ventilatoria entre sujetos de diferentes disciplinas deportivas, con diferentes niveles de entrenamiento, evaluados con diferentes ergómetros y con diferentes características antropométricas (**estudio IV**).
5- La eficiencia ventilatoria podría variar en condiciones de normoxia e hipoxia.	E. Conocer el comportamiento de le eficiencia ventilatoria en condiciones de hipoxia (FiO_2=16.45) y normoxia (FiO_2=21) (**estudio II**).	5) La respuesta de la eficiencia ventialtoria en condiciones de hipoxia (FiO_2=16.45) se ve modificada por la influencia de ésta en la ventilación (**estudio II**).
	J. Comparar el comportamiento de la eficiencia ventilatoria en condiciones de hipoxia y normoxia (**estudio II**).	10) La respuesta de la eficiencia ventilatoria durante el ejercicio se ve comprometida en situaciones ambientales de hipoxia (FiO_2=16.45) en comparación con las de normoxia (**estudio II**).
6- Un protocolo de entrenamiento de la musculatura inspiratoria podría promover o mejorar la respuesta de la eficiencia ventilatoria en deportistas.	H. Conocer si el entrenamiento de la musculatura inspiratoria mejora el rendimiento en condiciones de hipoxia (**estudio II**).	8) El entrenamiento de la musculatura inspiratoria mejora el rendimiento en condiciones de hipoxia (**estudio II**).
	F. Evaluar la influencia del entrenamiento de la musculatura respiratoria sobre la eficiencia ventilatoria de sujetos sanos (**estudio II**).	6) Un entrenamiento específico de los músculos inspiratorios no influye en sobre la respuesta de la eficiencia ventilatoria en condiciones normales de oxígeno (**estudio II**).
	G. Demostrar si el entrenamiento de la musculatura inspiratoria mejora la respuesta de la eficiencia ventilatoria en condiciones de hipoxia (**estudio II**).	7) El entrenamiento específico de los músculos inspiratorios podría tener un efecto positivo sobre la respuesta de la eficiencia ventilatoria en hipoxia (**estudio II**).
7- El entrenamiento interválico de alta intensidad podría provocar cambios en la eficiencia ventilatoria en deportistas.	L. Evidenciar si los posibles cambios en el rendimiento deportivo tras la aplicación de un programa de entrenamiento interválico de alta intensidad de 3 semanas están asociados o relacionados con una modificación de la eficiencia ventilatoria (**estudio III**).	12) Las pequeñas mejoras detectadas en sujetos entrenados tras un programa de entrenamiento interválico de alta intensidad no están asociadas a cambios o mejoras en la eficiencia ventilatoria (**estudio III**).

7- CONCLUSIONS [Conclusiones]

7.1. GENERAL CONCLUSION

The main conclusions obtained in this International PhD Thesis are:

1. Ventilatory efficiency might be a parameter which does not show intra-subject variability.

2. Neither un-controled training or specific training programs (IMT or HIT) might not promote changes on ventilatory efficiency response in athletes.

3. Ventilatory efficiency might be a parameter which react similarly in athletes independently of athlete´s characteristics or type of ergometer used for testing.

4. It might be possible that there is not relationship between ventilatory efficiency and sport performance.

7.2. SPECIFIC CONCLUSIONS RELATED TO THE AIMS

1) There is not relationship between the ability to eliminate CO_2 during exercise and sport performance (**publication I, II, III, IV**).

2) Ventilatory efficiency response is linked to the central respiratory control which does not allow intra-variability (**publication I, IV**).

3) Ventilatory efficiency reacted similarly in athletes independently of the fitness level, the age, the BMI or the ergometer used for testing (**publication IV**).

4) Ventilatory efficiency reacted similarly in sporty people (**publication I, II, III, IV**).

5) Ventilatory efficiency response is modified in hypoxia due to the influence of hypoxia (FiO_2=16.45) on ventilation (**publication II**).

6) Inspiratory muscle training did not influence ventilatory efficiency in normoxia (**publication II**).

7) Inspiratory muscle training could have a positive effect on ventilatory efficiency response in hypoxia (**publication II**).

8) Inspiratory muscle training improves cycling performance in hypoxia (**publication II**).

9) Improvements in sport performance after an inspiratory muscle training programme are not related to changes on VE/VCO_2 slope but are related to changes on OUES (**publication II**).

10) Ventilatory efficiency response is different under hypoxic conditions (FiO_2=16.45) to normoxia conditions (**publication II**).

11) Three weeks of high intensity-interval training did not change ventilatory efficiency response in well-trained athletes (**publication III**).

12) Changes in sport performance after three weeks high intensity-interval training of are not related to changes on ventilatory efficiency response in well-trained athletes (**publication III**).

HYPOTHESIS	AIMS	CONCLUSIONS
1- Ventilatory efficiency could be a sport performance variable in healthy people.	A. To know the relationship between ventilatory efficiency and sport performance (**publication I, II, III, IV**).	1) There is not relationship between the ability to eliminate CO_2 during exercise and sport performance **(publication I, II, III, IV).**
	K. To evaluate the ventilatory efficiency response after a training program (**studies II,III**).	11) Three weeks of high intensity-interval training did not changes ventilatory efficiency response in well-trained athletes **(publication III).**
		6) Inspiratory muscle training did not influence ventilatory efficiency in normoxia **(publication II).**
2- Changes in sport performance might be related to improvements on ventilator efficiency.	I. To know if changes in sport performance after an inspiratoy muscle training program could be related to changes on ventilatory efficiency (**publication II**).	9) Improvements in sport performance after an inspiratory muscle training program are not related to changes on VE/VCO_2 slope but are related to changes on OUES **(publication II).**
3- Changes in ventilatory efficiency response could be realted to changes in breathing pattern.	B. To understant the influence of central respiratory control on ventilatory efficiency response during exercise (**publication I, IV**).	2) Ventilatory efficiency response is linked to the central respiratory control which does not allow intra-variability (**publication I, IV**).
	D. To add information about ventilatory efficiency response during an incremental exercise test in athletes. (**estudios I, II, III, IV**).	4) Ventilatory efficiency reacted similarly in sporty people **(publication I, II, III, IV).**
4- Ventilatory efficiency migh response similar in athletes independently of their charactheristics.	C. To evaluate ventilatory efficiency in athletes with different fitness level, age, BMI and ergometer used for testing (**estudio IV**).	3) Ventilatory efficiency reacted similarly in athletes independently of the fitness level, the age, the BMI or the ergometer used for testing **(publication IV).**

5- Ventilatory efficiency may be different in normoxia and hipoxia.	E. To know ventilatory efficiency response under hypoxia (FiO_2=16.45) and normoxia (FiO_2=21) (**estudio II**).	5) Ventilatory efficiency response is modified in hypoxia due to the influence of hypoxia (FiO_2=16.45) on ventilation **(publication II).**
	J. To compare ventilatory efficiency in normoxia and hypoxia (**estudio II**).	10) Ventilatory efficiency response is different under hypoxic conditions (FiO_2=16.45) to normoxia conditions **(publication II).**
6- Inspiratory muscle training could improve ventilatory efficiency in athletes.	H. To know if inspiratory muscle training impoves sport performance in hypoxia (**estudio II**).	8) Inspiratory muscle training improves cycling performance in hypoxia **(publication II).**
	F. To evaluate the influence of an inspiratory muscle training on ventilatory efficiency (**estudio II**).	6) Inspiratory muscle training did not influence ventilatory efficiency in normoxia **(publication II).**
	G. To demostrate if an inspiratory muscle training improves ventilatory efficiency in hypoxia (**estudio II**).	7) Inspiratory muscle training could have a positive effect on ventilatory efficiency response in hypoxia **(publication II).**
7- A high-intensity interval training program could modify ventilatory efficiency in athletes.	L. To investigate if changes in sport performance after a three weeks of high-intensity inteval training are related to changes on ventialtory efficiency well-trained athletes (**estudio III**).	12) Changes in sport performance after three weeks high intensity-interval training of are not related to changes on ventilatory efficiency response in well-trained athletes **(publication III).**

OTRAS APORTACIONES / CONCLUSIONS

SALAZAR-MARTÍNEZ E.

OTRAS APORTACIONES CIENTÍFICAS [Other contributions]

Fruto del desarrollo de esta Tesis Doctoral Internacional se obtuvo otra contribución científica presentada en el XXI Congreso Europeo de Ciencias del Deporte (Viena). A continuación se exponen el abstract y la presentación de dicha comunicación en el libro de abstracts del congreso:

EFFECTS OF INSPIRATORY MUSCLE TRAINING ON VENTILATORY EFFICIENCY AND CYCLING PERFORMANCE

Introduction

It is well known that inspiratory muscle training (IMT) can improve performance in endurance athletes. However, the mechanisms responsible for these performance improvements remain unclear. Ventilatory efficiency might on factor potentially explaining these improvements (Sheel, 2002). The aim of this study was to investigate the influence of IMT on ventilatory efficiency and cycling performance.

Methods

Eight male and eight female subjects (VO_2max: 47.07± 5.64 $ml \cdot kg^{-1} \cdot min^{-1}$) were randomly assigned to the inspiratory muscles training group, (IMTG; n=9) or the control group (CG; n=9). Both groups performed an incremental cycle test to exhaustion with respiratory gas analysis, a peak inspiratory pressure test (Pimax) and performed a 10-min time trial (TT) before and after a 6-week lasting inspiratory muscle training period. The IMTG performed 60 IMT sessions, consisting of 30 breathing repetitions with a PowerBreathe® device at 50%Pimax, 2 times per day, 5 days per week. The control group performed the same breathing training without applying any resistance. The Pimax and training load was checked each week.

Results

Pimax, TT and peak power output (PPO) improved significantly within the IMTG ($p<0.05$). The VE/VCO_2 slope, ventilatory equivalent of CO_2 (VE/VCO_2) and VO_2max remained unchanged after the 6-week training period in both groups. Interaction effect was found between groups (group x time) in Pimax and TT after IMT. Significant correlations were found between Pimax, TT and PPO in both groups ($r=0.869$, $p<0.001$; $r=0.873$, $p<0.001$).

Discussion

Results are in accordance with previous work which demonstrated that IMT improved time trial cycling performance (Romer, McConnell, & Jones, 2002). However, our findings do not confirm the hypothesis that these improvements would be related to changes in ventilatory efficiency. Apparently the IMT is not able to modify the control of breathing and thus promoting more efficient breathing patterns. The sensitivity of the peripheral chemoreceptors seems not to be modified by the improvements in breathing muscle strength after IMT. Unchanged VE/VCO_2 values at the second ventilatory threshold indicate that the mechanical ventilatory efficiency was unaffected after IMT. Therefore, it is conclude that improvements in cycling time trial performance are not related to changes in ventilatory efficiency. However, our results suggested that the improvement in cycling performance might be related to improvements in breathing muscle strength.

REFERENCES

Romer, L. M., McConnell, A. K., & Jones, D. A. (2002). Inspiratory muscle fatigue in trained cyclists: effects of inspiratory muscle training. Medicine and science in sports and exercise, 34(5), 785-792.

Sheel, A. W. (2002). Respiratory muscle training in healthy individuals. Sports Medicine, 32(9), 567-581.

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BOOK OF ABSTRACTS

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Using crank angle obtained from the measurement system, the reacting forces of the bicycle and EMG signals were averaged at crank angle (every 5 degree) at each pedaling condition. Results & discussions: Using the averaged value of pedal force and pedaling action data vary with crank angle, inverse dynamics calculations have been performed by commercially available software (AnyBody). The saddle and handle force was not used for calculations, because it was difficult to take in. Since the freedom of pedaling motion remains, it is necessary to define the constraint condition of pedaling motion in the calculation. Moreover, calculation result was sometimes become unstable. As a result, calculated muscle activity changes can be compared with measured EMG values. However, these values did not match so well. Acknowledgments: This research was supported in part by JSPS KAKENHI Grant-in-Aid for Scientific Research (C), 25350763. References: (1) Tomoki Kitawaki & Hisao Oka A measurement system for the bicycle crank angle using a wireless motion sensor attached to the crank arm J Sci Cycling. Vol. 2(2), 13-19 Contact kitawaki@hirakata.kmu.ac.jp

EFFECTS OF INSPIRATORY MUSCLE TRAINING ON VENTILATORY EFFICIENCY AND CYCLING PERFORMANCE

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Introduction It is well known that inspiratory muscle training (IMT) can improve performance in endurance athletes. However, the mechanisms responsible for these performance improvements remain unclear. Ventilatory efficiency might on factor potentially explaining these improvements (Sheel, 2002). The aim of this study was to investigate the influence of IMT on ventilatory efficiency and cycling performance. Methods Eight male and eight female subjects (VO2max; 47.07± 5.64 ml·kg-1·min-1) were randomly assigned to the inspiratory muscles training group, (IMTG, n=9) or the control group (CG, n=9). Both groups performed an incremental cycle test to exhaustion with respiratory gas analysis, a peak inspiratory pressure test (Pimax) and performed a 10-min time trial (TT) before and after a 6-week lasting inspiratory muscle training period. The IMTG performed 60 IMT sessions, consisting of 30 breathing repetitions with a PowerBreathe® device at 50%Pimax, 2 times per day, 5 days per week. The control group performed the same breathing training without applying any resistance. The Pimax and training load was checked each week. Results Pimax, TT and peak power output (PPO) improved significantly within the IMTG ($p<0.05$). The VE/VCO2 slope, ventilatory equivalent of CO2 (VE/VCO2) and VO2max remained unchanged after the 6-week training period in both groups. Interaction effect was found between groups (group x time) in Pimax and TT after IMT. Significant correlations were found between Pimax, TT and PPO in both groups ($r=0.869$, $p<0.001$; $r=0.873$, $p<0.001$). Discussion Results are in accordance with previous work which demonstrated that IMT improved time trial cycling performance (Romer, McConnell, & Jones, 2002). However, our findings do not confirm the hypothesis that these improvements would be related to changes in ventilatory efficiency. Apparently the IMT is not able to modify the control of breathing and thus promoting more efficient breathing patterns. The sensitivity of the peripheral chemoreceptors seems not to be modified by the improvements in breathing muscle strength after IMT. Unchanged VE/VCO2 values at the second ventilatory threshold indicate that the mechanical ventilatory efficiency was unaffected after IMT. Therefore, it is conclude that improvements in cycling time trial performance are not related to changes in ventilatory efficiency. However, our results suggested that the improvement in cycling performance might be related to improvements in breathing muscle strength. References Romer, L. M., McConnell, A. K., & Jones, D. A. (2002). Inspiratory muscle fatigue in trained cyclists: effects of inspiratory muscle training. Medicine and science in sports and exercise, 34(5), 785-792. Sheel, A. W. (2002). Respiratory muscle training in healthy individuals. Sports Medicine, 32(9), 567-581.

CEREBRAL OXYGENATION DURING GRADED CYCLING TO EXHAUSTION IN WOMEN USING ORAL CONTRACEPTIVES

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Introduction Reduced maximal oxygen uptake (VO2max) has been reported in women using oral contraception (WomenOC) when compared to normally-menstruating women (WomenNM; Casazza et al. 2002; Joyce et al. 2013). A reduction in cerebral oxygenation during severe-intensity exercise has been associated with exercise exhaustion (Oussaidene et al. 2013) and may pose as a potential mechanism for the reduced VO2max observed in WomenOC. The aim of the present study was to investigate the effects of long-term use of oral contraceptives on frontal cortex oxygenation (i.e., cerebral oxygenation) during graded cycling to exhaustion in recreationally-active women. Methods Eight WomenOC and eight WomenNM performed a graded-cycling test to exhaustion to determine VO2max (VO2max test) and lactate threshold 1 and 2 (LT1 and LT2). Left pre-frontal cortex oxygenation was monitored by near-infrared spectroscopy (NIRS) through concentration changes in oxy-, deoxy-, and total haemoglobin (HbO2, HHb and tHb, respectively), along with an index of pre-frontal tissue saturation (TSI). Pulmonary gas exchange was measured throughout the VO2max test using open circuit spirometry. Results VO2max was not different between the two groups (WomenNM: 2.67(0.56) L/min; WomenOC: 2.69(0.47) L/min; $p = 0.83$). HbO2 was lower in WomenOC at rest; yet, the rate and magnitude of increase in HbO2 from the onset of exercise to LT1 were greater in WomenOC compared to WomenNM. At a work rate equal to that achieved at LT2, TSI declined for both groups, and all measures of cerebral oxygenation were similar between groups at exhaustion ($p>0.05$) Discussion When matched for VO2max, we observed different patterns of increase in cerebral oxygenation during graded cycling to exhaustion in WomenOC compared to WomenNM. However, there was no evidence of impaired cerebral oxygenation during severe-intensity exercise in either group. The suppression of 17β-estradiol and progesterone, as well as the elevation of ethinyl-estradiol and progestin associated with oral contraceptive use does not appear to affect cerebral hemodynamics during graded cycling. References Casazza, G., Suh S., Miller, B., Navazio, F., & Brooks, G. (2002). J Appl Physiol, 93, 1698-1702. Joyce, S., Sabapathy, S., Bulmer, A., & Minahan, C. (2013). J Strength Cond Res, 27(7), 1891-1896 Mendelsohn, M. (2002). Am J Cardiol, 90(1) F3-F4 Oussaidene, K., Prieur, F., Bougault, V., Borel, B., Matram, R., & Mucci, P. (2013). Eru J Appl Physiol, 113(8), 2047-2056
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HOW ACCESS TO AN E-BIKE AFFECTS AMOUNT AND PATTERNS OF BICYCLE USE IN INACTIVE NORWEGIAN ADULTS: A PILOT STUDY.

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Introduction Electric assisted bicycles (E-bikes) have become increasingly popular worldwide during the last decade. Bicycle use with an E-bike (E-biking) may be categorized as a physical activity of moderate intensity. Few studies have examined whether access to an E-bike influence bicycle use and fitness. The aims of the present study were to assess the effect of an eight-month intervention with access to an E-bike on (1) the amount of E-biking and (2) assess whether E-biking is associated with changes in maximal oxygen uptake (VO2 max),

REFERENCIAS / REFERENCES

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REFERENCIAS [References]

Akkerman, M., van Brussel, M., Hulzebos, E., Vanhees, L., Helders, P. J. M., & Takken, T. (2010). The oxygen uptake efficiency slope: what do we know? *Journal of cardiopulmonary rehabilitation and prevention, 30*(6), 357-373

Arena, Guazzi, & Myers. (2007). Prognostic value of end-tidal carbon dioxide during exercise testing in heart failure. *Int J Cardiol, 117*(1), 103-108. doi: 10.1016/j.ijcard.2006.04.058

Arena, Myers, Hsu, Peberdy, Pinkstaff, Bensimhon, . . . Guazzi. (2007). The minute ventilation/carbon dioxide production slope is prognostically superior to the oxygen uptake efficiency slope. *J Card Fail, 13*(6), 462-469. doi: 10.1016/j.cardfail.2007.03.004

Baba, R., Nagashima, M., Goto, M., Nagano, Y., Yokota, M., Tauchi, N., & Nishibata, K. (1996). Oxygen uptake efficiency slope: a new index of cardiorespiratory functional reserve derived from the relation between oxygen uptake and minute ventilation during incremental exercise. *Journal of the American College of Cardiology, 28*(6), 1567-1572

Barstow, T. J., Jones, A. M., Nguyen, P. H., & Casaburi, R. (2000). Influence of muscle fibre type and fitness on the oxygen uptake/power output slope during incremental exercise in humans. *Experimental physiology, 85*(1), 109-116

Benchetrit, G. (2000). Breathing pattern in humans: diversity and individuality. *Respiration physiology, 122*(2), 123-129

Bernardi, L., Schneider, A., Pomidori, L., Paolucci, E., & Cogo, A. (2006). Hypoxic ventilatory response in successful extreme altitude climbers. *European Respiratory Journal, 27*(1), 165-171

Brown, Raman, Schlader, & Stannard. (2013). Ventilatory efficiency in juvenile elite cyclists. *J Sci Med Sport, 16*(3), 266-270. doi: 10.1016/j.jsams.2012.06.010

Cardozo, G. G., Oliveira, R. B., & Farinatti, P. T. V. (2015). Effects of High Intensity Interval versus Moderate Continuous Training on Markers of Ventilatory and

Cardiac Efficiency in Coronary Heart Disease Patients. *The Scientific World Journal, 2015*

Coast, J. R., & Welch, H. G. (1985). Linear increase in optimal pedal rate with increased power output in cycle ergometry. *European journal of applied physiology and occupational physiology, 53*(4), 339-342

Coyle, E. F. (2005). Improved muscular efficiency displayed as Tour de France champion matures. *Journal of Applied Physiology, 98*(6), 2191-2196

Coyle, E. F., Sidossis, L. S., Horowitz, J. F., & Beltz, J. D. (1992). Cycling efficiency is related to the percentage of type I muscle fibers. *Medicine and science in sports and exercise, 24*(7), 782-788

Davis, Tyminski, Soriano, Dorado, Costello, Sorrentino, & Pham. (2006). Exercise test mode dependency for ventilatory efficiency in women but not men. *Clinical physiology and functional imaging, 26*(2), 72-78

Dempsey, J. A., Adams, L., Ainsworth, D. M., Fregosi, R. F., Gallagher, C. G., Guz, A., . . . Powers, S. K. (1996). Airway, lung, and respiratory muscle function during exercise. *Comprehensive Physiology*

Esposito, F., Limonta, E., Alberti, G., Veicsteinas, A., & Ferretti, G. (2010). Effect of respiratory muscle training on maximum aerobic power in normoxia and hypoxia. *Respiratory physiology & neurobiology, 170*(3), 268-272

Ettema, G., & Lorås, H. W. (2009). Efficiency in cycling: a review. *European journal of applied physiology, 106*(1), 1-14

Gaesser, G. A., & Brooks, G. A. (1975). Muscular efficiency during steady-rate exercise: effects of speed and work rate. *Journal of applied physiology, 38*(6), 1132-1139

Gilbert, C. (2005). Better chemistry through breathing: The story of carbon dioxide and how it can go wrong. *Biofeedback, 33*(3), 100-104

Gonzales, J. U., Tucker, S. H., Kalasky, M. J., & Proctor, D. N. (2012). Leg strength is associated with ventilatory efficiency in older women. *International journal of sports medicine, 33*(7), 537

Guerrero, L., Naranjo, J., & Carranza, M. D. (2008). Influence of gender on ventilatory efficiency during exercise in young children. *J Sports Sci, 26*(13), 1455-1457. doi: 10.1080/02640410802255771

Habedank, D., Reindl, I., Vietzke, G., Bauer, U., Sperfeld, A., Gläser, S., . . . Kleber, F. X. (1998). Ventilatory efficiency and exercise tolerance in 101 healthy volunteers. *European journal of applied physiology and occupational physiology, 77*(5), 421-426

Hall, J. E., & Guyton, A. C. (2001). Tratado de fisiologia médica. *Decima Primera Edicion*

Hopker, J. G., Coleman, D. A., & Wiles, J. D. (2007). Differences in efficiency between trained and recreational cyclists. *Applied Physiology, Nutrition, and Metabolism, 32*(6), 1036-1042

Illi, S. K., Held, U., Frank, I., & Spengler, C. M. (2012). Effect of respiratory muscle training on exercise performance in healthy individuals. *Sports medicine, 42*(8), 707-724

Ingle, L., Goode, K., Carroll, S., Sloan, R., Boyes, C., Cleland, J. G., & Clark, A. L. (2007). Prognostic value of the VE/VCO2 slope calculated from different time intervals in patients with suspected heart failure. *Int J Cardiol, 118*(3), 350-355. doi: 10.1016/j.ijcard.2006.07.105

Lucia, Carvajal, Calderon, Alfonso, & Chicharro. (1999). Breathing pattern in highly competitive cyclists during incremental exercise. *Eur J Appl Physiol Occup Physiol, 79*(6), 512-521. doi: 10.1007/s004210050546

Lucia, Hoyos, Pardo, & Chicharro. (2001). Effects of endurance training on the breathing pattern of professional cyclists. *Jpn J Physiol, 51*(2), 133-141

Lucia, Hoyos, Santalla, Earnest, & Chicharro. (2003). Giro, Tour, and Vuelta in the same season. *British journal of sports medicine, 37*(5), 457-459

Lucia, Hoyos, J., Pérez, M., Santalla, A., & Chicharro, J. L. (2002). Inverse relationship between VO2max and economy/efficiency in world-class cyclists. *Medicine & Science in Sports & Exercise, 34*(12), 2079-2084

Lucia, Pardo, Durantez, Hoyos, & Chicharro. (1998). Physiological differences between professional and elite road cyclists. *International journal of sports medicine, 19*(05), 342-348

Magrì, D., Limongelli, G., Re, F., Agostoni, P., Zachara, E., Correale, M., . . . Pacileo, G. (2016). Cardiopulmonary exercise test and sudden cardiac death risk in hypertrophic cardiomyopathy. *Heart*, heartjnl-2015-308453

McConnell, A. K., & Romer, L. M. (2004). Respiratory muscle training in healthy humans: resolving the controversy

McDaniel, J., Durstine, J. L., Hand, G. A., & Martin, J. C. (2002). Determinants of metabolic cost during submaximal cycling. *Journal of Applied Physiology, 93*(3), 823-828

Milic-Emili, G., & Cajani, F. (1957). Frequency of breathing as a function of respiratory ventilation during rest. *Bollettino della Società italiana di biologia sperimentale, 33*(6), 821-825

Milic-Emili, J. (1982). Recent advances in clinical assessment of control of breathing. *Lung, 160*(1), 1-17. doi: 10.1007/BF02719267

Milic-Emili, J., & Grunstein, M. M. (1976). Drive and timing components of ventilation. *Chest, 70*(1 Suppl), 131-133

Myers, J., Gademan, M., Brunner, K., Kottman, W., Boesch, C., & Dubach, P. (2012). Effects of high-intensity training on indices of ventilatory efficiency in chronic heart failure. *Journal of cardiopulmonary rehabilitation and prevention, 32*(1), 9-16

Palau, P., Domínguez, E., Núñez, E., Schmid, J.-P., Vergara, P., Ramón, J. M., . . . Núñez, J. (2014). Effects of inspiratory muscle training in patients with heart failure with preserved ejection fraction. *European journal of preventive cardiology, 21*(12), 1465-1473

Rodríguez, L. P., López-Rego, J., Calbet, J. A. L., Valero, R., Varela, E., & Ponce, J. (2002). Effects of training status on fibers of the musculus vastus lateralis in professional road cyclists. *American journal of physical medicine & rehabilitation, 81*(9), 651-660

Romer, L. M., McConnell, A. K., & Jones, D. A. (2002). Inspiratory muscle fatigue in trained cyclists: effects of inspiratory muscle training. *Medicine and science in sports and exercise, 34*(5), 785-792

Sallet, P., Mathieu, R., Fenech, G., & Baverel, G. (2006). Physiological differences of elite and professional road cyclists related to competition level and rider specialization. *Journal of sports medicine and physical fitness, 46*(3), 361

Santalla, A., Naranjo, J., & Terrados, N. (2009). Muscle efficiency improves over time in world-class cyclists. *Med Sci Sports Exerc, 41*(5), 1096-1101. doi: 10.1249/MSS.0b013e318191c802

Scheuermann, B. W., & Kowalchuk, J. M. (1999). Breathing patterns during slow and fast ramp exercise in man. *Exp Physiol, 84*(1), 109-120

Schneider, D. A., & Berwick, J. P. (1997). V˙ E and V˙ CO 2 remain tightly coupled during incremental cycling performed after a bout of high-intensity exercise. *European journal of applied physiology and occupational physiology, 77*(1), 72-76

Sheel, A. W. (2002). Respiratory muscle training in healthy individuals. *Sports Medicine, 32*(9), 567-581

Sidossis, L. S., Horowitz, J. F., & Coyle, E. F. (1992). Load and velocity of contraction influence gross and delta mechanical efficiency. *International journal of sports medicine, 13*(5), 407-411

Stainbsy, W. N., Gladden, L. B., Barclay, J. K., & Wilson, B. A. (1980). Exercise efficiency: validity of base-line subtractions. *Journal of applied physiology, 48*(3), 518-522

Sun, Hansen, Garatachea, Storer, & Wasserman. (2002). Ventilatory efficiency during exercise in healthy subjects. *American journal of respiratory and critical care medicine, 166*(11), 1443-1448

Terkelsen, K. E., Clark, A. L., & Hillis, W. S. (1999). Ventilatory response to erect and supine exercise. *Medicine and science in sports and exercise, 31*(10), 1429-1432

Tong, T. K., Fu, F. H., Chung, P. K., Eston, R., Lu, K., Quach, B., . . . So, R. (2008). The effect of inspiratory muscle training on high-intensity, intermittent running performance to exhaustion. *Applied physiology, nutrition, and metabolism, 33*(4), 671-681

Tortora, G., & Derrickson, B. (2007). *Principios de anatomía y fisiología*: Panamericana.

Ukkonen, H., Burwash, I. G., Dafoe, W., Kemp, R. A., Haddad, H., Yoshinaga, K., . . . Beanlands, R. S. B. (2008). Is ventilatory efficiency (VE/VCO2 slope) associated with right ventricular oxidative metabolism in patients with congestive heart failure? *European journal of heart failure, 10*(11), 1117-1122

Zavin, A., Arena, R., Joseph, J., Allsup, K., Daniels, K., Schulze, P. C., . . . Forman, D. E. (2013). Dynamic assessment of ventilatory efficiency during recovery from peak exercise to enhance cardiopulmonary exercise testing. *European journal of preventive cardiology, 20*(5), 779-785

ANEXOS / ATTACHED DOCUMENTS

SALAZAR-MARTÍNEZ E.

ANEXOS [Attached documents]

ÍNDICE DE ANEXOS

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Ventilatory efficiency and breathing pattern in world-class cyclists: A three-year observational study

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ABSTRACT

The purpose of this three-year observational study was to analyze the ventilatory efficiency and breathing pattern in world-class professional cyclists. Twelve athletes (22.61 ± 3.8 years; 177.38 ± 5.5 cm; 68.96 ± 5.5 kg and VO_{2max} 75.51 ± 3.3mL kg^{-1} min^{-1}) were analyzed retrospectively. For each subject, respiratory and performance variables were recorded during incremental spiroergometry: oxygen uptake (VO_2), carbon dioxide output (VCO_2), pulmonary ventilation (VE), tidal volume (Vt), breathing frequency (f_R), driving (Vt/Ti), timing (Ti/Ttot), peak power output (PPO) and maximum oxygen uptake (VO_{2max}). Ventilatory efficiency (VE/VCO_2 slope) was calculated from the beginning of exercise testing to the second ventilatory threshold (VT_2). The VE/VCO_2 slope was unaffected during the study period (24.63 ± 3.07; 23.61 ± 2:79; 24:89 ± 2:61) with a low effect size (ES = 0.04). The PPO improved significantly in the third year (365 ± 33.74; 386.36 ± 32.33; 415.00 ± 24.15) ($p < 0.05$). The breathing pattern variables, Vt/Ti and Ti/Ttot, did not change significantly over the three year period (ES = 0.00; ES = 0.03 respectively). These findings suggest that changes in cycling performance in world-class professional cyclists do not modify breathing variables related to the control of ventilatory efficiency.

1. Introduction

Carbon dioxide (CO_2) is produced in cellular metabolism and expelled into the atmosphere by ventilation (VE), but during this process CO_2 plays a fundamental role in the regulation of bodily pH, vascular tone (Gilbert, 2005) and in the ventilation control (Milsom et al., 2004). The relationship between the rate of CO_2 output (VCO_2) and VE in different circumstances has been widely described as a measurement of breathing efficiency (Arena et al., 2007b; Arena et al., 2008; Sun et al., 2002) at a given metabolic rate. During incremental effort, the slope of the linear relationship between VE and VCO_2 (VE/VCO_2 slope or DeltaCO_2) is the most widely used method to evaluate ventilatory efficiency (Arena et al., 2007b; Brown et al., 2013; Schneider and Berwick, 1998; Sun et al., 2002; Ukkonen et al., 2008).

The VE/VCO_2 slope has been commonly used in patients suffering from congestive heart failure (Arena et al., 2007a,b; Ingle et al., 2007; Laveneziana et al., 2010; Robertson, 2011) as well as in healthy subjects (Sun et al., 2002). It is well established that the values of the VE/VCO_2 slope vary from 19 to 32 in healthy subjects (Sun et al., 2002), with values exceeding 34 considered abnormal (Arena et al., 2007a,b) or indicative of the inefficiency of the respiratory system (Brown et al., 2013). The large variability in VE/VCO_2 slope could be an inborn characteristic but it could also be explained by the lack of consensus of measurement methods. Thus, differences in the VE/VCO_2 slope arise depending on whether it is measured from rest to VT_2 or from rest to the maximal work load.

In trained athletes, the VE/VCO_2 slope has not been widely studied and its relationship with sport performance is unclear. It could be possible that two athletes had different values of equivalent of CO_2 (VE/VCO_2) to the same metabolic rate, but they show the same VE/VCO_2 slope throughout the entire incremental test. In this case, they have different efficiency to a given level but the same global efficiency because they need the same increase in VE for every increase of 1 l min^{-1} in CO_2 production (VE/VCO_2 slope) during the incremental test. It could be possible that the high demands of elite cycling promote a lower VE/VCO_2 slope, involving a lower increase in VE for a given increment in VCO_2. Conditions where the CO_2 production is elevated, such as cycling, seems to play an essential role in the ventilatory control (Milsom et al., 2004). The ventilatory efficiency control could change over time in presence of a large amount of training and competition as it happens with

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others respiratory and performance variables (Lucia et al., 1998; Sallet et al., 2006).

Accepted that the VE/VCO_2 slope in normal subjects is a marker of ventilatory sensitivity, there are three possible mechanisms by which respiratory efficiency could change with training. One is through changes in the dead space (VD) at low power output (Wood et al., 2008); the other is a better mechanical performance of respiratory muscles (Sheel, 2002) and the third is an improved sensitivity of chemoreceptors (Babb et al., 2010).

It may also be expected that changes in the VE/VCO_2 slope with training could be related to improvements of breathing control by a more effective breathing pattern. Unlike ventilatory efficiency, the breathing pattern has been widely studied in athletes (Benchetrit, 2000; Lucia et al., 1999; Lucia et al., 2001; Scheuermann and Kowalchuk, 1999). A simple way to analyze the breathing pattern is to evaluate the relationship between the tidal volume (Vt) and breathing frequency (f_R) (Milic-Emili and Cajani, 1957). However, since the 1970s (Milic-Emili, 1982; Milic-Emili and Grunstein, 1976), it has been known that VE can be decomposed into the product of two components which offer more information: (a) central inspiratory activity, known as "driving" and expressed as the relationship between Vt and inspiratory time (Vt/Ti) and (b) the inspiration-expiration alternation, known as "timing", and expressed by the relationship between Ti and the total duration of the breathing cycle (Ti/Ttot). The analysis of all these variables (VE, Vt, BF, Vt/Ti, Ti/Ttot) is nowadays the most widely-used method to evaluate the breathing pattern in patients (Beltrão et al., 2015), sedentary subjects (Benchetrit, 2000) and athletes as well (Lucia et al., 2001; Lucia et al., 2003).

The studies which evaluated the VE/VCO_2 slope in healthy people were performed with sedentary or moderately trained subjects but not in highly trained athletes. In addition, we are not aware of any articles dealing with longitudinal VE/VCO_2 slope observations in athletes. It is usually difficult to study cyclists of the highest level due to the number of hours of training and competition they undergo over the competitive seasons, but thanks to the evaluations carried out by our research group over several seasons with a UCI-Pro Tour team (Santalla et al., 2009), we were able to analyze all these respiratory variables of interest in world-class cyclists.

We hypothesized that the high demands of professional cycling could induce changes in ventilatory efficiency, measured as the VE/VCO_2 slope, in world-class cyclists over a three-year period. If true, we would expect that changes in the VE/VCO_2 slope are related to changes in breathing pattern.

Therefore, the purpose of this study was to perform a retrospective longitudinal evaluation of the ventilatory efficiency and breathing pattern in world-class cyclists.

2. Material and methods

2.1. Subjects

A total of 42 male world-class professional cyclists, tested during the same period of the season in the same laboratory for at least five seasons, were retrospectively analyzed to select those for whom consecutive evaluations were available over a period of at least three years. Finally, 12 cyclists (starting age 22.61 ± 3.8 years; height 177.38 ± 5.5 cm; body weight 68.96 ± 5.5 kg and 76.92 ± 5.9 mL kg^{-1} min^{-1}) were included who had participatedannually in at least one of the three-week stage races (Giro d'Italia, Tour de France and Vuelta a España) or were evaluated at least two times consecutively over three years at the same time point in the season. Some of the subjects were among the best cyclists in the world (one winner of the Tour de France, one winner of the Vuelta a España and first in the annual ICU world ranking, one three-time Tour de France Podium, two Vuelta a España Podium, one Junior World Time Trial Champion, and two one-week stage race winners). All subjects provided written informed consent before testing. The study has been approved by the ethics committee of the Pablo de Olavide University (Seville).

2.2. Exercise test

Tests were always conducted during the first phase of the cyclists' competitive season. All incremental exercise tests have been performed on the same electromagnetically braked cycle ergometer. This ergometer allowed the subjects to choose their own pedal frequency and to adopt a position similar to that on their bicycles (Orion S.T.E., Toulouse, France). The distances and dimensions for saddle, handlebars and connecting rod were monitored and remained constant during the entire test period. The test was started at a power output (PO) of 100 W, after which PO was increased by 50 W every 4 min until exhaustion. This exercise protocol has already been used in previous research (Fernandez-Garcia et al., 2000). The freely chosen pedaling cadence generally ranged from 77 to 115 revolutions per min (rpm). Heart rate was monitored using radio-telemetry (Sport tester PE 4000; Polar, Kempele, Finland). Ventilation and respiratory gases were measured continuously and the highest 30-s VO_2 value was considered as VO_{2max}. VO_2, VCO_2, VE, Vt, f_R, Ti, Te and Ttot were measured breath by breath (BxB) using a gas analyzer (Vmax 29; Sensormedics, Yorba Linda, CA), which was calibrated before every exercise session. During the data collection period (three years), neither the cycle ergometer nor the gas analyzer was replaced and all the equipment passed the maintenance procedures recommended by the manufacturers. The ergometer was calibrated by the manufacturers annually. In addition, all the tests were performed under similar ambient temperature conditions (20–24C and 45%–65% relative humidity).

2.3. Ventilatory efficiency and breathing pattern

The ventilatory efficiency of each subject was calculated from the slope of the relationship between VCO_2 and VE during each test. To exclude the influence due to respiratory compensation for acidosis during highly intensive exercise, the VE/VCO_2 slope was determined from the beginning of the test until the second ventilatory threshold (VT_2). The breathing pattern was evaluated by the analysis of Vt, f_R, Vt/Ti and Ti/Ttot. The value of the slope representing the relationship between VE and Vt/Ti during each test (the driving slope) was used to test the central component.

2.4. Statistical analysis

The data is expressed as mean ± SD for each variable. The normality of the data was checked by means of the Shapiro-Wilk test. The homogeneity of variance was evaluated by Leveneís test. To compare the values obtained for each variable during the three-year observation period, one-way ANOVA with repeated measurements and the Friedman F-test (nonparametric conditions) were used. When significant differences were found, the Bonferroni test was used as a post hoc test. Effect sizes (ES) were also calculated using Eta-Squared. Intra-class correlations (ICC) and Pearson correlation coefficient (Pearson-r) were used to determine the reproducibility of measurements over time for VE/VCO_2 slope, VO_{2max} and PPO.Correlation analyses were carried out between VO_{2max}, VE/VCO_2 slope and PPO. The level of significance was set at $p < 0.05$ for each statistical analysis.

Table 1
Evolution of ventilatory and performance variables during the study period.

	1st year	2nd year	3th year	*p*-value	Effect Size
VE/VCO_2 slope	24.63 ± 3.07	23.61 ± 2.79	24.89 ± 2.61	0.549	0.04
VT/Ti slope	30.79 ± 1.59	31.09 ± 1.29	30.86 ± 1.38	0.883	0.00
Vt/Ti (l seg^{-1}) $_{(uptoPPO)}$	5.13 ± 0.29	5.17 ± 0.53	5.02 ± 0.47	0.541	0.02
Ti/Ttot $_{(uptoPPO)}$	0.47 ± 0.01	0.46 ± 0.02	0.47 ± 0.01	0.655	0.03
Vt (l) $_{(uptoPPO)}$	3.15 ± 0.55	3.14 ± 0.46	3.28 ± 0.55	0.694	0.01
Vt (l) $_{(uptoVT2)}$	2.7 ± 469	2.84 ± 398	2.86 ± 428	0.659	0.02
f_R (breaths min^{-1}) $_{(uptoPPO)}$	50.50 ± 10.36	50.64 ± 10.13	48.90 ± 11.10	0.457	0.00
f_R (breaths min^{-1}) $_{(uptoVT2)}$	35 ± 5.77	37 ± 7.31	35.2 ± 5.79	0.73	0.02
VO_{2max} (mL kg min^{-1})	75.92 ± 6.28	75.29 ± 6.09	77.93 ± 5.31	0.578	0.03
VE_{max} (l min^{-1})	155.54 ± 17.97	156.37 ± 17.76	153.33 ± 15.98	0.812	0.00
HR_{max} (beats min^{-1})	183.2 ± 8.33	183.4 ± 8.98	184.4 ± 7.76	0.539	0.00
PPO (W)	365 ± 33.74	386.36 ± 32.33	415.00 ± 24.15	0.011*	0.32*

VE/VCO_2 slope, ventilatory efficiency; Vt/Ti slope, driving; Ti/Ttot, timing; Vt, tidal volume; f_R breathing frequency; VO_{2max}, maximum oxygen uptake, VE_{max}, maximum ventilation, HR_{max}, maximum heart rate PPO, peak power output. All values are expressed as mean ± SD.
*Significantly different from 1st year vs 3th year in PPO ($p < 0.05$).
*Large effect size (ES ≥ 0.14).

Table 2
Test-re-test reliability between repeated measurements over three years study period.

	CCI	Pearson r 1st year–2nd year	1st year–3th year	2nd year–3th year
VE/VCO_2 slope	0.896	0.900*	0.911*	0.745*
VO_{2max}(mL·kg·min^{-1})	0.234	0.543	−0.392	0.234
PPO (W)	0.029	0.688*	0.745*	0.378

CCI, correlation coefficient intra-class; VE/VCO_2 slope, ventilatory efficiency; VO_{2max}, maximum oxygen uptake, PPO, peak power output.
* Significantly correlation ($p < 0.05$).

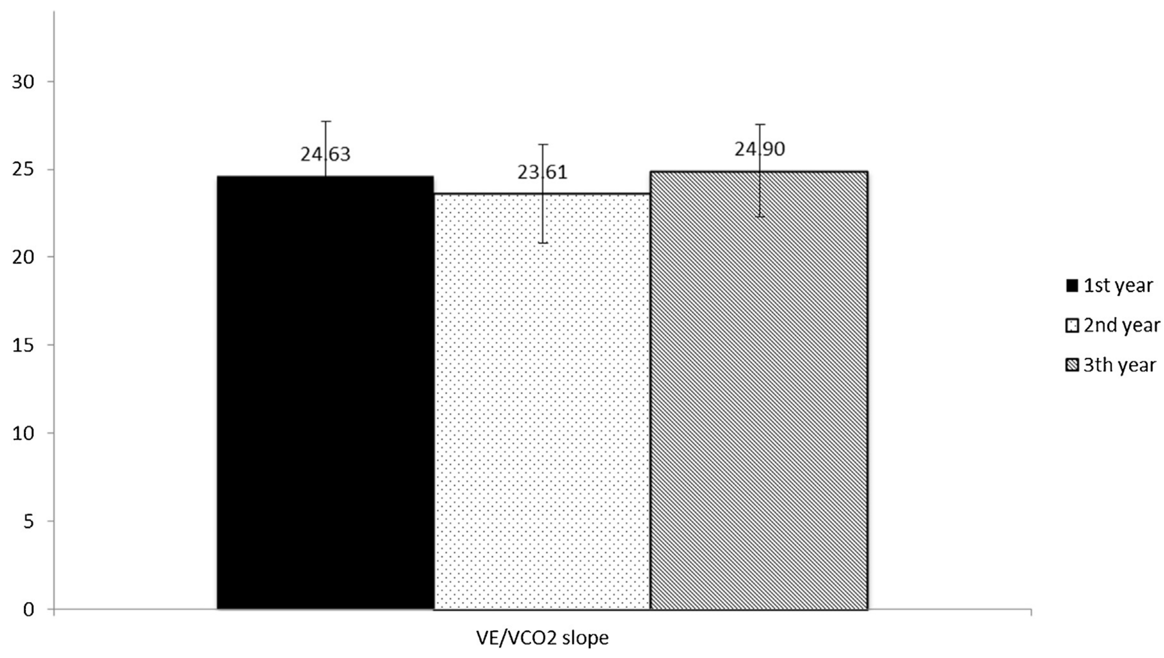

Fig. 1. Means values of ventilatory efficiency (VE/VCO_2 slope) between years in world-class cyclists. No significant differences were found between years.

3. Results

Data on the ventilatory and performance variables evaluated are shown in Table 1. No significant differences were found in any of the respiratory variables studied. VE/VCO_2 slope and VO_{2max} show a weak effect size (ES = 0.04; 0.03) respectively. Significant differences were found in the PPO between the first and the third year, whit a large effect size (ES = 0.32). Table 2 shows the test-re-test reliability, with the ICC and Pearson-r for VE/VCO_2 slope, VO_{2max} and PPO measurements. Figs. 1, 2 and 3 show the mean values obtained each year for VE/VCO_2 slope, VO_{2max} and PPO respectively. Fig. 4 shows the correlation between VCO_2 and VE for each year. The overall value of VE/VCO_2 slopes obtained was similar to the means values found each year. Figs. 5 and 6 demonstrates the relationship between VO_{2max}, VE/VCO_2 slope and PPO with the values of all subjects over the entire study period. No correlations were found between VE/VCO_2 slope and VO_{2max} and between VE/VCO_2 slope and PPO.

4. Discussion

To the best of our knowledge, this is the first longitudinal study which analyzed the ventilatory efficiency (VE/VCO_2 slope) and breathing pattern in world-class cyclists. The main finding of the

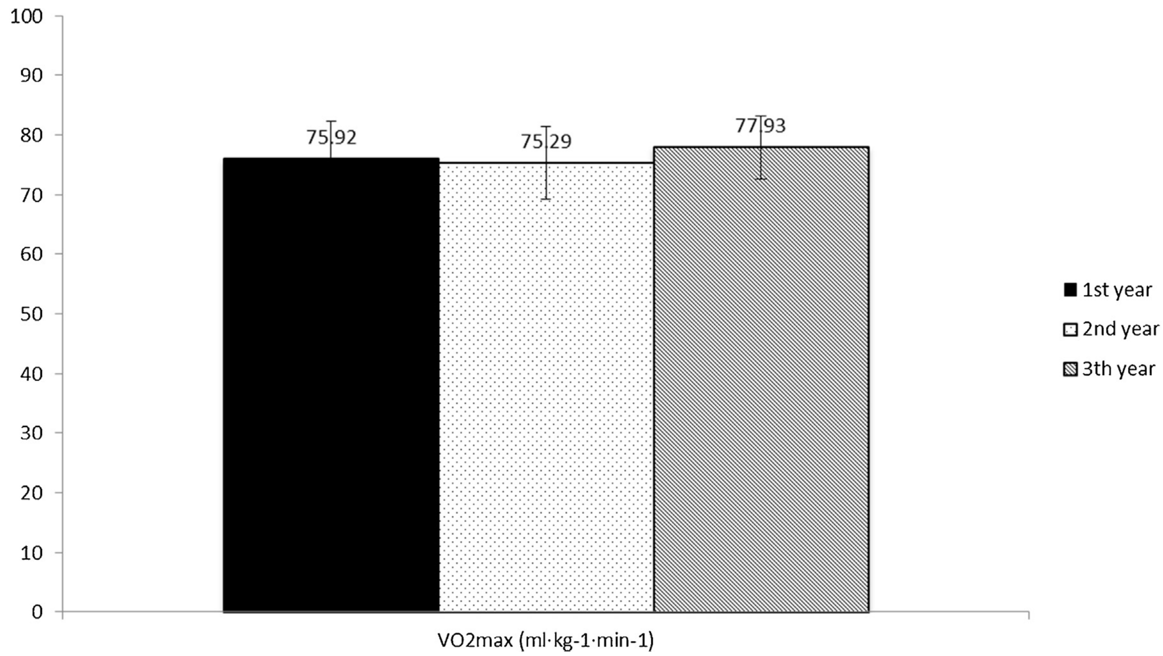

Fig. 2. Means values of maximum oxygen uptake (VO_{2max}) over three years study period in world-class cyclists. No significant differences were found between years.

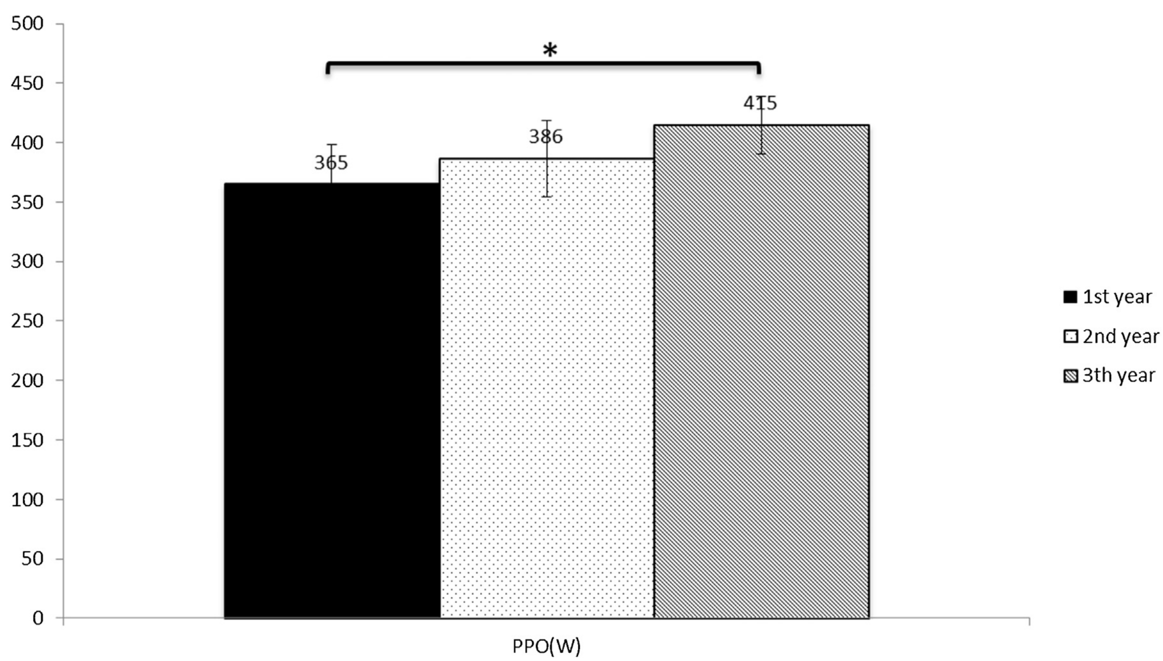

Fig. 3. Means values of peak power output (PPO) over three years study period in world-class cyclists. Significant differences were found between 1st and 3th year ($p < 0.05$).

present study was that ventilatory efficiency, measured as VE/VCO_2 slope, and breathing pattern did not change in top cyclists over a three-year observation period. Our results agree with previous data suggesting that ventilatory efficiency would be a variable which is maintained within a constant range regardless of physical effort and training adaptations (Brown et al., 2013; Guerrero et al., 2008; Sun et al., 2002; Terkelsen et al., 1999).

Why efficiency could change over time in cyclists? Three possibilities could be involved: (1) changes in the dead space (VD); (2) better mechanical performance of respiratory muscles; (3) changes in sensitivity of carotic bodies. Given that this study was retrospective, we could not choose variables to measure. But, not finding any change in VE/VCO_2 slope, we can assume that none of these variables had to change or at least have not conditioned changes in respiratory efficiency.

Concerning dead space, we know from experiments with goats (Mitchell, 1990) that ventilation is stimulated by anadded dead space and this seems to be so in healthy humans during exercise (Wood et al., 2008) resulting in an increased VE/VCO_2 slope, implying a lower ventilatory efficiency. However, as the exercise intensity increased, the effect of the added dead space was reduced (Babb et al., 2010). This effect of added space is considered a short term modulation (STM) (Babb et al., 2010), but there is no evidence to support ventilatory control during exercise being influenced by hyperpnoeic history (long term modulation, LTM) associated with

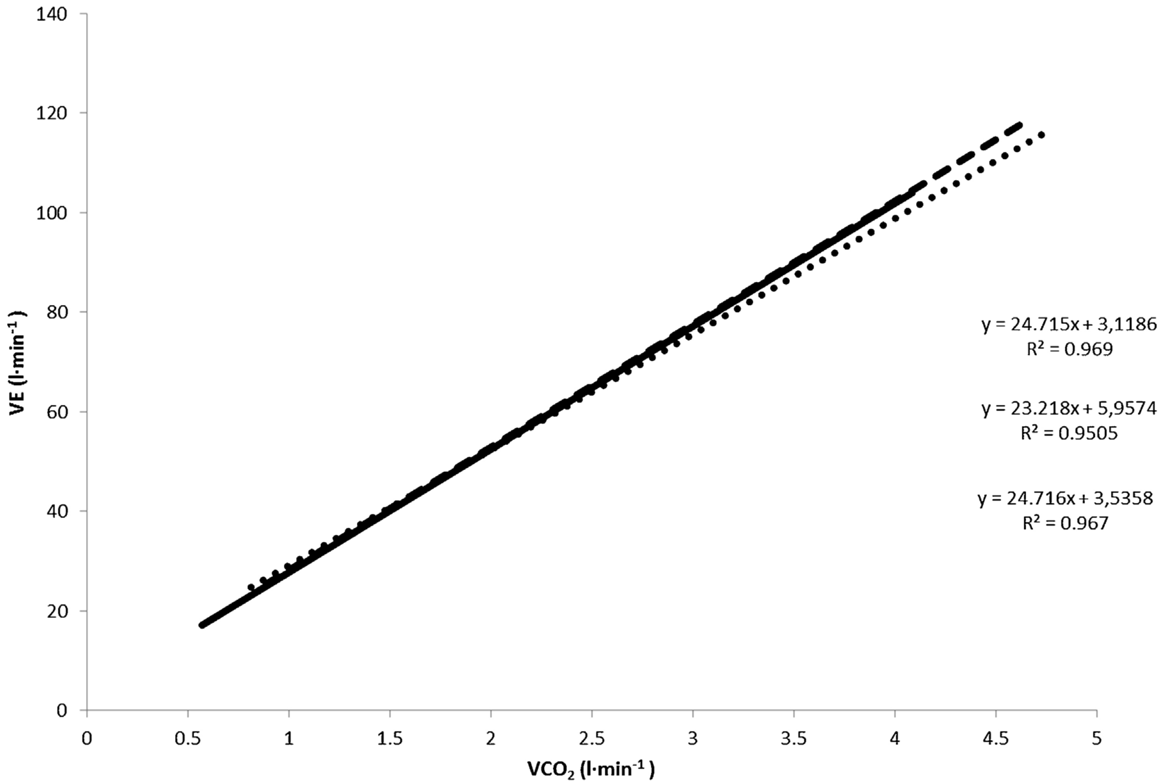

Fig. 4. Relationship between ventilation (VE) and CO_2 output (VCO_2) showing values measured each year over the three-year observation period.

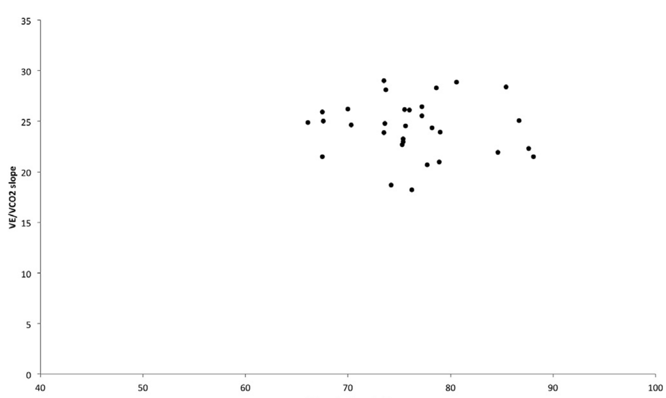

Fig. 5. Relationship between ventilatory efficiency (VE/VCO_2 slope) and maximum oxygen uptake (VO_{2max}) over three years study period in world-class cyclists. No relationship was found between VE/VCO_2 slope and VO_{2max}.

dead-space loading in humans (Cathcart et al., 2005) and even less in high-level athletes.

With respect to the mechanical performance of respiratory muscles, studies carried out with world-class professional cyclists, both transversal (Lucia et al., 2002) and longitudinal (Coyle, 2005; Hopker et al., 2010; Santalla et al., 2009; Sassi et al., 2008), found evidence that training increases muscular and mechanical performance. An inverse correlation between VO_{2max} and gross efficiency (GE) calculated as the ratio of work acomplished min^{-1} (i.e. W converted to Kcal min^{-1}), has been reported in world-class professional cyclists (Lucia et al., 2002). The authors suggested that this is a potential predictor of performance in professional cycling since the subjects with a higher GE were those with better results in time trials and in the overall standings in the Giro, Tour and Vuelta. In the same way, previous results obtained by our research group have described an increase in cycling efficiency in world-class professional cyclists, suggesting that this increase could even be a strategy to compensate a lower VO_{2max}, thereby maintaining a high competitive level (Santalla et al., 2009). Some studies suggest that specific respiratory muscle training can improve the endurance and strength of the respiratory muscles in healthy humans, although the effects on exercise performance remain controversial (Sheel, 2002). Therefore, it would be possible that a better trained inspiratory muscles contributes to more efficient ventilation.

Regarding sensitivity of chemoreceptors, we could not analyze it in a retrospective study, but we agree with other authors (Mitchell, 1990; Babb et al., 2010) that respiratory efficiency is not related to changes in chemoreflex stimulation.

Anyway, our results indicate that the training adaptations in high level athletes which cause changes in performance (Fig. 3) and other efficiency variables (Lucia et al., 2002; Santalla et al., 2009) do not modify ventilatory efficiency. The test-re-test reliability for VE/VCO_2 slope measurements was high (CCI = 0.89), with a great coefficient of determination (all ≥0.8), this help to add evidence that the VE/VCO_2 slope remains unchanged over time. These subjects constantly increase their VE by approximately 24 L min^{-1} for each L min^{-1} of increase in VCO_2 during incremental cycling ergometry (Table 1). These values are indicative of high ventilatory efficiency, in fact they are lower than values found in juvenile cyclists (∼28) (Brown et al., 2013). Based on the few papers that have studied this variable during exercise, it seems that the VE/VCO_2 slope tends to remain constant, at least in healthy subjects (Sun et al., 2002). No significant differences in ventilatory efficiency have been found after 16-weeks of training (Brown et al., 2013), during exercise between sexes (Guerrero et al., 2008), age groups (Sun et al., 2002) or bodily position (Terkelsen et al., 1999).

Concerning the breathing pattern, both Vt/Ti and Ti/Ttot behave similarly in healthy people when ventilation is stimulated, regardless of the stimulus (exercise, CO_2 inhalation, etc.)(McConnell and Davies, 1992; Szekely et al., 1982).During progressive treadmill exercise, it is known thatTi/Ttot remained nearly constant while

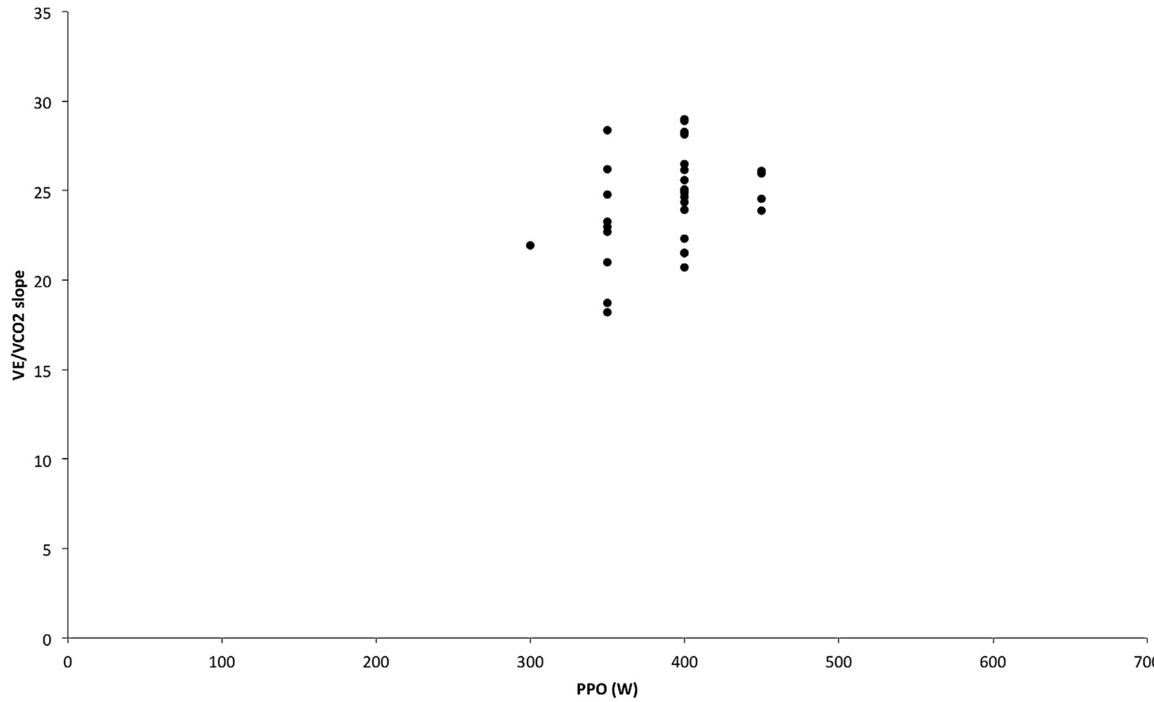

Fig. 6. Relationship between ventilatory efficiency (VE/VCO_2 slope) and peak power output (PPO) over three years study period in world-class cyclists. No relationship was found between VE/VCO_2 slope and PPO.

Vt/Ti increased linearly with VE and that this response was independent of gender and protocol (ramp or step) (Naranjo et al., 2005). However, only a few studies evaluated the ventilatory response to exercise in elite cyclists over time and most of them were limited to only analysis of changes in VE (Hoogeveen, 2000). Lucia et al. (1999) analyzed the breathing pattern in a cross-sectional study with highly competitive cyclists during incremental exercise, comparing amateur and professional cyclists. They demonstrated that Vt/Ti and Ti/Ttot showed a similar response in both groups. In addition, Scheuermann and Kowalchuk (1999) showed that Ti/Ttot and Vt/Ti were similar during a slow ramp ($8\,W\,min^{-1}$) and fast ramp protocol ($65\,W\,min^{-1}$) at all submaximal exercise intensities, suggesting that breathing pattern and respiratory timing may behave independently of alveolar and arterial PCO_2. Recently, Tanner et al. (2014) showed that there are no differences between cycling and running in breathing pattern variablesat maximal exercise intensities. There has been also reported an inter-individual variability in breathing patterns at high levels of exercise, suggesting that respiratory control by peripheral vagal afferents seems to prevail in subjects who increase f_R and not Vt after VT_2 (Gravier et al., 2013). In our study the breathing patter of athletes remain unchanged. Ti/Ttot values remained without significant change throughout the different tests (Table 1), while Vt/Ti values showed an almost perfect linear relationship with VE values (Fig. 2). This indicates that the increases in VE during progressive exercise are associated with a proportional increase in the inspiratory driving activity without any alteration in the relationship between inspiration and expiration, even at the highest working intensities (over 400 W). Although this coincides with previous transversal evidence (Lucia et al., 1999, 2001; Naranjo et al., 2005), it is the first time to be described longitudinally over a period of more than one season.

With respect to the variables related to performance, the stability of VO_{2max} observed (Fig. 2) over the three seasons and the increase in PPO (Fig. 3) agree with the previous research with world-class cyclists (Lucia et al., 2002; Santalla et al., 2009). Correlations analysis performed suggested that there is no relationship between the cycling performance and ventilatory efficiency in world-class cyclists (Figs. 5 and 6).

At least three limitations have to be addressed. First, we do not have exact data on the amount of training and competitions completed by the cyclists. However, due to the inclusion criteria, we strongly assume that all of them trained and competed similarly. Second, the sample size is relatively small but should be sufficiently large considering the known difficulties of gaining access to world-class cyclists. Thirdly, this study was retrospective and we could not choose the variables to measure; for example, we could not measure arterial PCO_2.

New research to evaluate the influence of specific training program on ventilatory efficiency and VA in athletes is necessary in order to better clarify the involvement and influence of this variable on ventilation performance during exercise. It would be useful to include measures of PaCO2 and VD in new designs.

5. Conclusions

In conclusion, the presented findings do not support our hypothesis that performance changes in world-class cyclists over a three-year period would be associated with changes in ventilatory efficiency and breathing pattern. They rather suggest that the central control of respiration, responsible for promoting efficient breathing patterns in response to exercise moves within very tight ranges, is set by the system and was not modified through training and competition. Furthermore, the efficiency of the CO_2 elimination during exercise also appears to be preserved and closely related to the central control mechanism of ventilation.

References

Arena, R., Guazzi, M., Myers, J., 2007a. Prognostic value of end-tidal carbon dioxide during exercise testing in heart failure. Int. J. Cardiol. 117, 103–108.

Arena, R., Myers, J., Hsu, L., Peberdy, M.A., Pinkstaff, S., Bensimhon, D., Chase, P., Vicenzi, M., Guazzi, M., 2007b. The minute ventilation/carbon dioxide production slope is prognostically superior to the oxygen uptake efficiency slope. J. Card. Fail. 13, 462–469.

Arena, R., Myers, J., Guazzi, M., 2008. The clinical and research applications of aerobic capacity and ventilatory efficiency in heart failure: an evidence-based review. Heart. Fail. Rev. 13, 245–269.
Babb, T.G., Wood, H.E., Mitchell, G.S., 2010. Short- and long-term modulation of the exercise ventilatory response. Med. Sci. Sports Exerc. 42, 1681–1687.
Beltrão, B.A., Herdman, T.H., Pascoal, L.M., Chaves, D.B.R., Silva, V.M., Lopes, M.V., 2015. Ineffective breathing pattern in children and adolescents with congenital heart disease: accuracy of defining characteristics. J. Clin. Nurs. 24 (17–18), 2505–2513.
Benchetrit, G., 2000. Breathing pattern in humans: diversity and individuality. Respir. Physiol. 122, 123–129.
Brown, S.J., Raman, A., Schlader, Z., Stannard, S.R., 2013. Ventilatory efficiency in juvenile elite cyclists. J. Sci. Med. Sport 16, 266–270.
Cathcart, A.J., Herrold, N., Turner, A.P., Wilson, J., Ward, S.A., 2005. Absence of long-term modulation of ventilation by dead-space loading during moderate exercise in humans. Eur. J. Appl. Physiol. 93, 411–420.
Coyle, E.F., 2005. Improved muscular efficiency displayed as Tour de France champion matures. J. Appl. Physiol. (Bethesda Md.:1985) 98, 2191–2196.
Fernandez-Garcia, B., Perez-Landaluce, J., Rodriguez-Alonso, M., Terrados, N., 2000. Intensity of exercise during road race pro-cycling competition. Med. Sci. Sports Exerc. 32, 1002–1006.
Gilbert, C., 2005. Better chemistry through breathing: the story of carbon dioxide and how it can go wrong. Biofeedback 33, 100–104.
Gravier, G., Delliaux, S., Delpierre, S., Guieu, R., Jammes, Y., 2013. Inter-individual differences in breathing pattern at high levels of incremental cycling exercise in healthy subjects. Respir. Physiol. Neurobiol 189, 59–66.
Guerrero, L., Naranjo, J., Carranza, M.D., 2008. Influence of gender on ventilatory efficiency during exercise in young children. J. Sports Sci. 26, 1455–1457.
Hoogeveen, A.R., 2000. The effect of endurance training on the ventilatory response to exercise in elite cyclists. Eur. J. Appl. Physiol. 82, 45–51.
Hopker, J., Coleman, D., Passfield, L., Wiles, J., 2010. The effect of training volume and intensity on competitive cyclists' efficiency. Appl. Physiol. Nutr. Metab. 35, 17–22.
Ingle, L., Goode, K., Carroll, S., Sloan, R., Boyes, C., Cleland, J.G., Clark, A.L., 2007. Prognostic value of the VE/VCO2 slope calculated from different time intervals in patients with suspected heart failure. Int. J. Cardiol. 118, 350–355.
Laveneziana, P., Agostoni, P., Mignatti, A., Mushtaq, S., Colombo, P., Sims, D., Uriel, N., Jorde, U.P., 2010. Effect of acute β-blocker withholding on ventilatory efficiency in patients with advanced chronic heart failure. J. Card. Fail. 16 (7), 548–555.
Lucia, A., Pardo, J., Durantez, A., Hoyos, J., Chicharro, J.L., 1998. Physiological differences between professional and elite road cyclists. Int. J. Sports Med. 19, 342–348.
Lucia, A., Carvajal, A., Calderon, F.J., Alfonso, A., Chicharro, J.L., 1999. Breathing pattern in highly competitive cyclists during incremental exercise. Eur. J. Appl. Physiol. Occup. Physiol. 79, 512–521.
Lucia, A., Hoyos, J., Pardo, J., Chicharro, J.L., 2001. Effects of endurance training on the breathing pattern of professional cyclists Jpn. J. Physiol. 51, 133–141.
Lucia, A., Hoyos, J., Pérez, M., Santalla, A., Chicharro, J.L., 2002. Inverse relationship between VO2max and economy/efficiency in world-class cyclists. Med. Sci. Sports Exerc. 34, 2079–2084.
Lucia, A., Hoyos, J., Santalla, A., Earnest, C.P., Chicharro, J.L., 2003. Giro Tour, and Vuelta in the same season. Br. J. Sports Med. 37, 457–459.
McConnell, A.K., Davies, C.T., 1992. A comparison of the ventilatory responses to exercise of elderly and younger humans. J. Gerontol. 47, B137–141.
Milic-Emili, G., Cajani, F., 1957. [Frequency of breathing as a function of respiratory ventilation during rest]. Boll. Soc. Ital. Biol. Sper. 33, 821–825.
Milic-Emili, J., Grunstein, M.M., 1976. Drive and timing components of ventilation. Chest 70, 131–133.
Milic-Emili, J., 1982. Recent advances in clinical assessment of control of breathing. Lung 160, 1–17.
Milsom, W.K., Abe, A.S., Andrade, D.V., Tattersall, G.J., 2004. Evolutionary trends in airway CO_2/H^+ chemoreception. Respir. Physiol. Neurobiol. 144, 191–202.
Mitchell, G.S., 1990. Ventilatory control during exercise with increased respiratory dead space in goats. J. Appl. Physiol. 69, 718–727.
Naranjo, J., Centeno, R.A., Galiano, D., Beaus, M., 2005. A nomogram for assessment of breathing patterns during treadmill exercise. Br. J. Sports Med. 39, 80–83.
Robertson, H.T., 2011. Gas Exchange consequences of left heart failure. Compr. Physiol. 1, 621–634.
Sallet, P., Mathieu, R., Fenech, G., Baverel, G., 2006. Physiological differences of elite and professional road cyclists related to competition level and rider specialization. J. Sports Med. Phys. Fitness 46, 361–365.
Santalla, A., Naranjo, J., Terrados, N., 2009. Muscle efficiency improves over time in world-class cyclists. Med. Sci. Sports Exerc. 41, 1096–1101.
Sassi, A., Impellizzeri, F.M., Morelli, A., Menaspa, P., Rampinini, E., 2008. Seasonal changes in aerobic fitness indices in elite cyclists. Appl. Physiol. Nutr. Metab. 33, 735–742.
Scheuermann, B.W., Kowalchuk, J.M., 1999. Breathing patterns during slow and fast ramp exercise in man. Exp. Physiol. 84, 109–120.
Schneider, D.A., Berwick, J.P., 1998. VE and VCO2 remain tightly coupled during incremental cycling performed after a bout of high-intensity exercise. Eur. J. Appl. Physiol. Occup. Physiol 77, 72–76.
Sheel, A.W., 2002. Respiratory muscle training in healthy individuals: physiological rationale and implications for exercise performance. Sports Med. 32, 567–581.
Sun, X.G., Hansen, J.E., Garatachea, N., Storer, T.W., Wasserman, K., 2002. Ventilatory efficiency during exercise in healthy subjects. Am. J. Respir. Crit. Care Med. 166, 1443–1448.
Szekely, O., Askanazi, J., Rosenbaum, S.H., Foster, R., Milic-Emili, J., Kinney, J.M., 1982. Ventilatory patterns during steady state and progressive exercise. Clin. Physiol. 2, 391–400.
Tanner, D.A., Duke, J.W., Stager, J.M., 2014. Ventilatory patterns differ between maximal running and cycling. Respir. Physiol. Neurobiol. 191, 9–16.
Terkelsen, K.E., Clark, A.L., Hillis, W.S., 1999. Ventilatory response to erect and supine exercise. Med. Sci. Sports Exerc. 31, 1429–1432.
Ukkonen, H., Burwash, I.G., Dafoe, W., de Kemp, R.A., Haddad, H., Yoshinaga, K., Davies, R.A., Gannon, E.K., Dasilva, J.N., Beanlands, R.S., 2008. Is ventilatory efficiency (VE/VCO(2) slope) associated with right ventricular oxidative metabolism in patients with congestive heart failure? Eur. J. Heart Fail. 10, 1117–1122.
Wood, H.E., Mitchell, G.S., Babb, T.G., 2008. Short-term modulation of the exercise ventilatory response in young men. J. Appl. Physiol. 104, 244–252.

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Influence of Inspiratory Muscle Training on Ventilatory Efficiency and Cycling Performance in Normoxia and Hypoxia

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The aim of this study was to analyse the influence of inspiratory muscle training (IMT) on ventilatory efficiency, in normoxia and hypoxia, and to investigate the relationship between ventilatory efficiency and cycling performance. Sixteen sport students (23.05 $\pm$ 4.7 years; 175.11 $\pm$ 7.1 cm; 67.0 $\pm$ 19.4 kg; 46.4 $\pm$ 8.7 ml·kg^{-1}·min^{-1}) were randomly assigned to an inspiratory muscle training group (IMTG) and a control group (CG). The IMTG performed two training sessions/day [30 inspiratory breaths, 50% peak inspiratory pressure (Pimax), 5 days/week, 6-weeks]. Before and after the training period subjects carried out an incremental exercise test to exhaustion with gas analysis, lung function testing, and a cycling time trial test in hypoxia and normoxia. Simulated hypoxia ($FiO_2 = 16.45\%$), significantly altered the ventilatory efficiency response in all subjects ($p < 0.05$). Pimax increased significantly in the IMTG whereas no changes occurred in the CG (time × group, $p < 0.05$). Within group analyses showed that the IMTG improved ventilatory efficiency (V_E/VCO_2 slope; $EqCO_2VT_2$) in hypoxia ($p < 0.05$) and cycling time trial performance [$W_{TTmax\ (W)}$; $W_{TTmean\ (W)}$; $PTF_{(W)}$] ($p < 0.05$) in hypoxia and normoxia. Significant correlations were not found in hypoxia nor normoxia found between ventilatory efficiency parameters (V_E/VCO_2 slope; $LEqCO_2$; $EqCO_2VT_2$) and time trial performance. On the contrary the oxygen uptake efficiency slope (OUES) was highly correlated with cycling time trial performance ($r = 0.89$; $r = 0.82$; $p < 0.001$) under both conditions. Even though no interaction effect was found, the within group analysis may suggest that IMT reduces the negative effects of hypoxia on ventilatory efficiency. In addition, the data suggest that OUES plays an important role in submaximal cycling performance.

Keywords: V_E/VCO_2 **slope, cycling performance, ventilation, chemosensitivity, time trial**

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INTRODUCTION

Ventilatory efficiency can be defined as the relationship between carbon dioxide production (VCO_2) and ventilation (V_E). Increased V_E and the removal of CO_2 during physical exercise are essential for homeostatic control of whole body pH (Brown et al., 2013). There are four common ways for measuring ventilatory efficiency during an incremental test: (a) using the slope of the relationship between VCO_2 and V_E (V_E/VCO_2 slope; Ingle et al., 2007), (b) the lowest equivalent of

CO_2during the incremental test (LEqCO_2; Sun et al., 2002), (c) the equivalent of CO_2 at the second ventilatory threshold (Eq$CO_2$$VT_2$; Sun et al., 2002), and (d) the oxygen uptake efficiency slope (OUES; Baba et al., 1999a). Generally, a lower equivalent of CO_2 indicates a greater ventilatory efficiency (Sun et al., 2002). The OUES represents the rate of increase of VO_2 in response to a given V_E during incremental exercise, indicating how effectively oxygen is extracted and taken into the body (Baba et al., 1996).

In the clinic field ventilatory efficiency has been widely used as a prognostic marker to determine exercise limitation (Ingle et al., 2007; Arena et al., 2008; Laveneziana et al., 2010). Indeed, a relationship has been reported between sudden dead risk in hypertrophic cardiomyopathy and ventilatory efficiency (Magrì et al., 2016). However, the importance of the ventilatory efficiency for sport performance remains unclear. On one hand, Brown et al. (2013) did not find a relationship between maximum oxygen uptake (VO_{2max}) and OUES in juvenile cyclists. In the same way, we did not find a relationship between VO_{2max}, peak power output (PPO), and V_E/VCO_2 slope in world-class cyclists (Salazar-Martínez et al., 2016). On the other hand, a significant correlation was found between OUES and VO_{2max} in young active women (Mourot et al., 2004).

In hypoxia, the reduced partial pressure of oxygen (PO_2) and the resulting arterial desaturation stimulates V_E (Babcock et al., 1995). Although the increased V_E during exercise in hypoxia ($FIO_2 = 0.15$) increases PaO_2 (Warner and Mitchell, 1990) it also leads to a higher oxygen cost of breathing compared to normoxia (Babcock et al., 1995). Additionally, in hypoxia, V_E may increase in excess of what would be required to maintain partial pressure levels of carbon dioxide ($PaCO_2$; Warner and Mitchell, 1990). Therefore, a certain ventilatory inefficiency could be expected in hypoxia due to this overshoot in V_E. However, high ventilatory efficiency is essential to maintain adequate level of PaO_2 and $PaCO_2$ with a lower breathing work in high altitude (Bernardi et al., 2006). In this regard, it may be assumed that efficient breathing may play an important role in the regulation of PaO_2 and $PaCO_2$ and achieving a higher sport performance in hypoxia.

In accordance, inspiratory muscle training (IMT) has been shown to be an effective method to improve both the ventilatory response in normoxia and hypoxia (Downey et al., 2007; Esposito et al., 2010) and the alveolar-arterial gradient in hypoxia (Esposito et al., 2010). Thus, it could be speculated that well-trained inspiratory muscles may help to preserve PaO_2 in hypoxia due to improved ventilation-perfusion matching and to prevent excessive CO_2 output due to less hyperventilation. However, to the best of our knowledge, whether or not IMT may improve the ventilatory efficiency under hypoxia conditions has not yet been tested.

Next to the effect on ventilation, IMT has been shown to improve sport performance as well (Romer et al., 2002a; Wells et al., 2005). However, the mechanisms responsible for the performance improvements after IMT remain controversial (Edwards and Walker, 2009). The mechanisms suggested to improve performance include a hypertrophy of diaphragm (Downey et al., 2007), an increase in blood flow to the locomotor muscles (Harms et al., 1997) and a reduction in subjective perception of fatigue and dyspnea ratings (Downey et al., 2007). Additionally, Sheel (2002) hypothesized that changes in performance after IMT could be related to improvements on ventilatory efficiency. However, to the best of our knowledge there are no studies evaluating the relationship between changes in sport performance after IMT and ventilatory efficiency. After IMT, the metabolic demand of the inspiratory muscles during exercise are reduced (Babcock et al., 1995), thus contributing to a lower overall O_2 uptake and CO_2 output. In situations where the ventilatory efficiency is impaired, for example in hypoxia, such effects may influence exercise performance (Roussos, 1985).

Therefore, the aim of this study was (a) to evaluate the influence of IMT on ventilatory efficiency in normoxia and hypoxia, and (b) to investigate the relationship between ventilatory efficiency and cycling performance under both conditions.

We hypothesized that IMT improves ventilatory efficiency in normoxia and especially in hypoxia and reduces the metabolic demands of the respiratory muscles in both conditions. We also hypothesized that improvements in submaximal cycling performance can be linked to improvements in ventilatory efficiency in normoxia and hypoxia.

MATERIALS AND METHODS

Subjects

Sixteen physically active and healthy participants [$n = 9$ male (23.44 ± 2.7 years; 180.22 ± 3.5 cm; 78.2 ± 5.5 kg; 48.39 ± 7.28 ml·kg^{-1}·min^{-1}); $n = 7$ female (25.37 ± 3.24 years; 168.75 ± 5.1 cm; 62.62.2 ± 9.47 kg; 38.15 ± 6.57 ml·kg^{-1}·min^{-1})] were selected for the study. Each participant completed a health questionnaire before being included in the study. Participants with health diseases, breathing problems, or obstructive defects were excluded from the study. Before starting the study, written informed consent was obtained from each participant in accordance with the Declaration of Helsinki. The study was approved by the Ethics Committee of the University of Innsbruck.

Design

Participants were randomly assigned to either an inspiratory muscle training group (IMTG) or a control group (CG). The IMTG performed two training sessions per day, 5 days per week during a period of 6-weeks. Each participant completed 30 inspiratory breaths with a PowerBreath device (PowerBreathe®, K3) at 50% of their individual Pimax. Inspiratory training load was adjusted weekly at 50% of the individual Pimax. Every training session was performed under expert supervision. The CG did not carry out any inspiratory training during the experimental period. This procedure seems to be adequate as a placebo effect is not expected. For instance, when considering differences between trials that included a control group and studies that did not, 69% of the placebo-controlled studies showed a positive outcome for RMT (i.e., performance improvements for the RMT groups significantly exceeded those for the control groups), which is very similar to the 75% positive outcomes of the studies without any controls (Illi et al., 2012).

Participants were advised not to change normal physical training habits during the experimental period.

Pulmonary Function Tests

Before and after the experimental period, participants performed lung function testing (Schiller SP-1®, Switzerland) to assess the forced vital capacity (FVC), forced expiratory volume during the first second (FEV_1), the ratio between forced expiratory capacity during the first second and vital capacity (FEV_1/VC), the peak expiratory flow (PEF), and the peak inspiratory flow (PIF; **Table 1**). The best attempt out of three tests was included in the analysis. Peak inspiratory mouth pressure (Pimax) was determined with a portable device (PowerBreathe®, K3). During the Pimax test participants had to inspire as fast as possible from a normal expiration. Each participant repeated the test until the measurements were stable. Pimax was measured weekly using the same testing protocol.

Incremental Exercise Testing

Before (Pre) and after (Post) the training period participants performed maximum incremental exercise tests in normoxia and hypoxia (overall four tests). Each test was separated by 48 h. During the tests, oxygen uptake (VO_2), carbon dioxide output (VCO_2), respiratory exchange ratio (RER), ventilation (V_E), breathing frequency (BF), tidal volume (VT), oxygen equivalent ($EqVO_2$), and carbon dioxide equivalent ($EqCO_2$) were measured breath by breath with a portable gas analyser (Jaeger Oxygen™®, Germany). The system was calibrated prior to each test with gas mixtures of known concentration. Tests were carried out on a cycle ergometer (RBM Cyclus 2®, Germany). After 4 min of warming up, participants started the test at 50 W and then the load was increased by 25 W each minute until volitional exhaustion. Achievement of maximum oxygen uptake (VO_{2max}) was accepted when a plateau was found in the relationship between VO_2 and power output or when three of the four criteria for maximal VO_{2max} were obtained (Howley et al., 1995). Tests were carried out at approximately the same time of the day in an air-conditioned normobaric hypoxic chamber (size 4.75 × 2.25 m, LowOxygen®, Germany). During the normoxia testing the hypoxic chamber was switched off whereas during the hypoxia setting the chamber was set at a simulated altitude of 2,500 m ($FiO_2 = 16.45\%$). Participants were blinded to the simulated altitude of the hypoxic chamber. Participants were advised to avoid exhausting exercise 1 day before the tests and to take any ergogenic aids (e.g., caffeine).

TABLE 1 | Results of pulmonary function testing pre and post experimental period (Mean ± *SD*).

	IMTG		CG	
	Pre	Post	Pre	Post
FVC (l)	5.44 ± 1.14	4.67 ± 1.38	5.06 ± 1.17	4.96 ± 0.93
FEV_1 (l)	4.64 ± 0.92	4.19 ± 0.8	4.31 ± 0.85	4.06 ± 0.79
FEV_1/VC (%)	84.13 ± 11.58	82.51 ± 9.19	82.33 ± 6.28	79.84 ± 6.48
PEF ($l \cdot s^{-1}$)	9.27 ± 2.23	8.2 ± 1.53	8.9 ± 2.47	8.73 ± 2.4
PIF ($l \cdot s^{-1}$)	7.04 ± 1.92	8.31 ± 2.39	7.12 ± 1.2	7.57 ± 2.2
Pimax (cm H_2O)	119.6 ± 37.36	166.91 ± 42.65*	130.55 ± 33.58	146.72 ± 40.62

FVC, forced vital capacity; FEV1, forced expiratory volume during the first second; FEV1/VC, ratio between forced expiratory volume during the first second and vital capacity; PEF, peak expiratory flow; PIF, peak inspiratory flow; Pimax, peak inspiratory pressure.

**$p < 0.05$ post vs. pre training.*

Time Trial Performance

Ninety minutes after the incremental test, cycling endurance performance was evaluated by a 10 min time trial (TT). The cycle ergometer was shifted to a fixed pedal force in which power output was dependent on the pedaling rate. Pedal force for each participant was set in order that pedaling at 90 rpm produced 85% (rounded to 5 W) of peak power output determined by the incremental cycle ergometer test. During the test, cyclists were strongly encouraged to choose a maximal pedaling rate that could be maintained for the respective test duration. As with the incremental test, each participant performed the TT, under normoxic (TT_{nor}) and under hypoxic conditions (TT_{hyp}) separated by 48 h, before and after the experimental period. During each TT-test peak power output (W_{max}), mean power output (W_{mean}), and pedal torque force (PTF) were recorded.

Test–Retest Reproducibly of Time Trial Test

The coefficient of variation for the time trial test in the control group in normoxia was 14.9% in the pre-test and 15.9% in the post-test (**Figure 1A**). In hypoxia, the coefficient of variation for the time trial test in the control group was 17.9% in the pre-test and 17.8% in the post-test (**Figure 1B**). The intra-class correlation coefficient for the time trial test in the control group was 0.92 in normoxia and 0.93 in hypoxia.

Ventilatory Efficiency

The V_E/VCO_2 slope was calculated from the slope of the relationship between VCO_2 and V_E during each incremental exercise test. To exclude the influence of the respiratory compensation due to acidosis during highly intensive exercise, the V_E/VCO_2 slope was determined from the beginning of the test until the second ventilatory threshold (VT_2). VT_2 was identified by an increase in the ventilatory equivalent of CO_2 ($EqCO_2$) and a decrease in end tidal partial pressure of carbon dioxide ($PETCO_2$; Lucía et al., 2000). Oxygen uptake efficiency slope (OUES) was calculated from the linear relationship of VO_2 vs. the logarithm of V_E during exercise ($VO_2 = a \log_{10} V_E + b$).

Statistics Analysis

Data are expressed as mean ± *SD* for each variable. The statistical power for the chosen sample size of 16 participants (9 in the IMTG and 7 in the CG) was >90%; alpha = 0.05. The power calculation (G*Power 3.1.7) was based on expected changes in Pimax and TT performance (W_{TTmean}) due to IMT (Romer et al., 2002a). The normal distribution of the data was checked by the

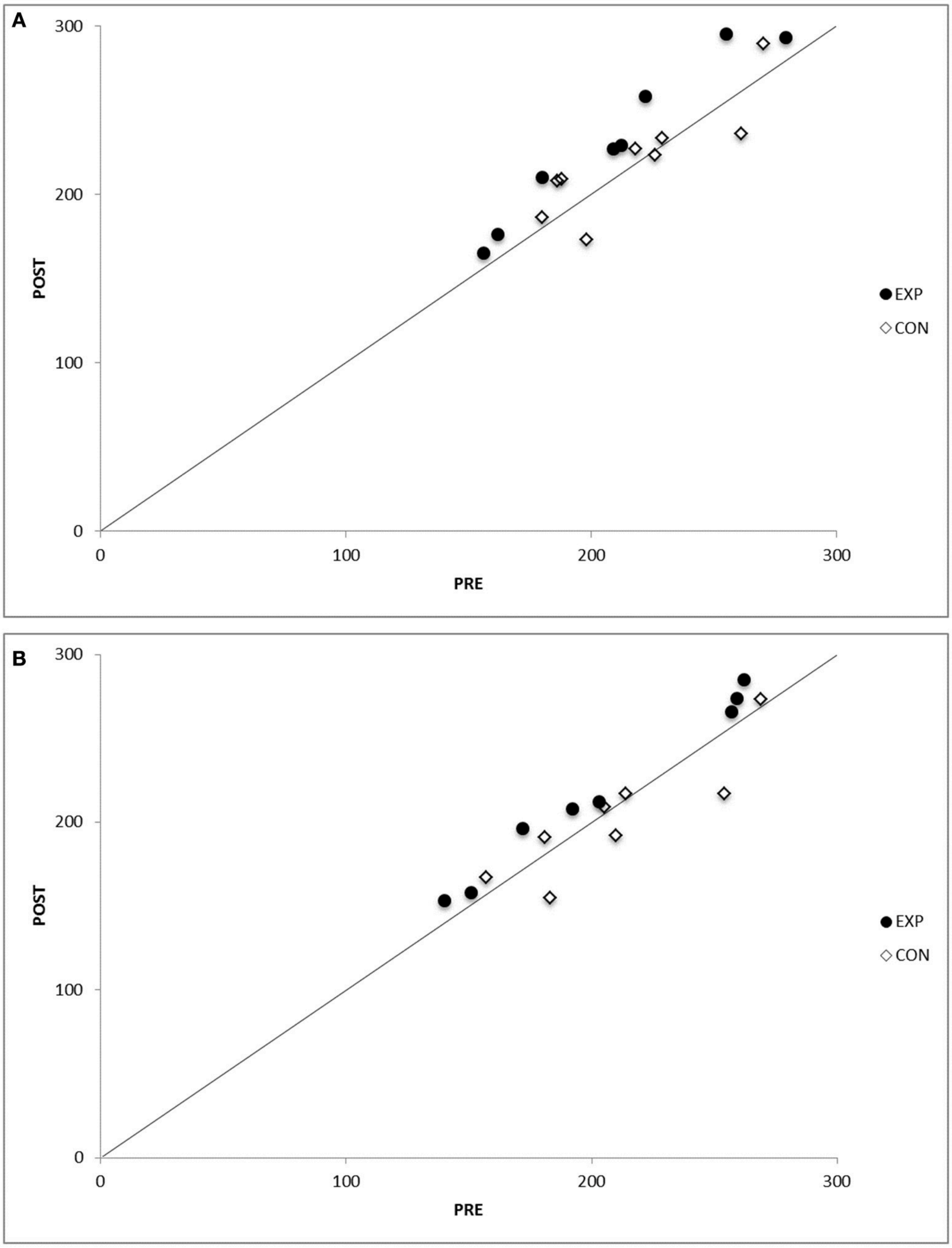

FIGURE 1 | (A) Test–retest reproducibility in the control/experimental group subjects during the time trial test (TT) test, before (Pre), and after (Post) the intervention period in normoxia. Identity lines are drawn in both graphs. See text for numerical analysis. **(B)** Test–retest reproducibility in the control/experimental group subjects during the time trial test (TT) test, before (Pre), and after (Post) the intervention period in hypoxia. Identity lines are drawn in both graphs. See text for numerical analysis.

Shapiro-Wilk test. The homogeneity of variance was evaluated by Levene's test. To compare the values obtained for each variable during the test, mixed-effects ANOVA test was used (group × time × condition). When significant differences were found, the Bonferroni test was used as a *post-hoc* test. ANOVA test was also applied to evaluate a possible gender effect (group × time × condition × gender). Effect size (ES) was calculated when a significant difference was found. A correlation analysis (Pearson-coefficient) was carried out between TT performance variables, incremental test variables and ventilatory efficiency variables with data from both groups and from both test in two different situations (normoxia and hypoxia; **Table 6**). Linear regression analysis was performed between Pimax, V_E/VCO_2 slope and OUES (dependent variables) and TT performance (independent variable) with data from both groups (IMTG and CG) and from both tests (Pre and Post) in normoxia. The level of significance was set at $p < 0.05$ for each statistical analysis.

RESULTS

No gender effect was identified with regard to the parameters of interest (V_E/VCO_2 slope, $LEqCO_2$, $EqCO_2VT_2$, OUES). Baseline values did not differ between groups (V_E/VCO_2 slope, $LEqCO_2$, $EqCO_2VT_2$, OUES, Pimax, PPO, $TT_{W\,mean}$). Outcomes of the pulmonary function testing before and after the experimental period are shown in **Table 1**. Significant differences were found in Pimax between Pre- and Post-test in the IMTG ($p < 0.05$).

Inspiratory Muscle Training (IMT)

Figure 2 shows the changes in Pimax during the experimental period in both groups. Mean Pimax improved significantly from week-1 to week-6 in the IMTG (+28.37%, $p < 0.05$) with no improvements in the CG (interaction effect, time × group, $p < 0.05$).

Ventilatory Efficiency

The group comparison of the ventilatory efficiency variables are shown in **Table 3**. There were no group differences and no interaction effect in any of the established variables at Pre and Post in normoxia and hypoxia. Significant differences were found between normoxia and hypoxia in the V_E/VCO_2 slope, $LEqCO_2$, and $EqCO_2VT_2$ in the Pre-test in both groups ($p < 0.05$; **Table 2**). During the Post-test significant differences between normoxia and hypoxia were found in $LEqCO_2$ and OUES in the IMTG and in V_E/VCO_2 slope, $LEqCO_2$ and OUES in the CG ($p < 0.05$). In both groups significant differences in V_E/VCO_2 slope and $EqCO_2VT_2$ were found between Pre and Post in hypoxia ($p < 0.05$).

Time Trial Performance

Time trial performance parameters are shown in **Table 4**. During Pre- and Post-test, significant differences between normoxia and hypoxia were found in $W_{TTmean\,(W)}$ and in $W_{TTmean\,(W/Kg)}$ in both groups ($p < 0.05$). There was no interaction effect in these variables. However, after the experimental period, $W_{TTmean\,(W)}$ and $W_{TTmean\,(W/Kg)}$ were significantly higher in normoxia and hypoxia in the IMTG ($p < 0.05$). At post, significant differences were found in W_{max} between normoxia and hypoxia in both groups ($p < 0.05$). A significant reduction in PTF was found in the CG in hypoxia in both tests (Pre and Post) and in the Post-test in the IMTG ($p < 0.05$). A significant increase in PTF was found in the IMTG in normoxia after IMT ($p < 0.05$).

Incremental Exercise Testing

VO_{2max} was reduced in hypoxia in both groups compared to normoxia in the Post-test ($p < 0.05$). Compared to normoxia, PPO was reduced in both groups during the Pre- and Post-test in hypoxia ($p < 0.05$). There was no interaction effect. However, PPO increased significantly in the IMTG in normoxia after the experimental period compared to the Pre-test evaluation ($p < 0.05$). Before the experimental period, V_{Emax} increased

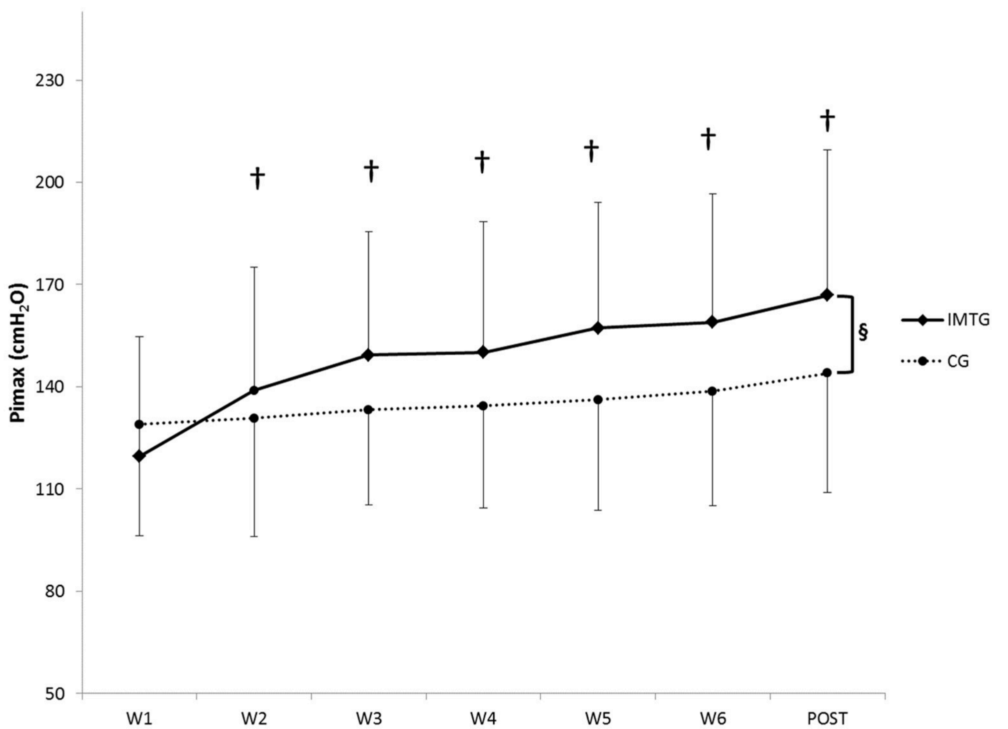

FIGURE 2 | Weekly values of Pimax (Mean ± *SD*) for the inspiratory muscle training group (IMTG) and control group (CG). [§]Two-way ANOVA for repeated measures (time × group) interaction ($p < 0.05$). [†]Differences from baseline evaluation (Bonferroni test).

in hypoxic conditions in both groups. After the experimental period, V_{Emax} increased only in the CG. However, all these variations were not significant in both gorups. VT_{max} and BF_{max} did not change in any condition.

Correlation and Regression Analysis

Significant correlations were found between Pimax and W_{TTmean}, VO_{2max}, V_{Emax}, and PPO with data from both test and both groups in normoxia ($p < 0.05$; **Table 6**). No correlation was found between ventilatory efficiency variables and performance variables. A significant correlation was found between OUES and maximal performance variables (VO_{2max}, V_{Emax}, PPO) and W_{TTmean} ($p < 0.05$) in normoxia and hypoxia (**Table 6**). A linear relationship was found between muscle breathing strength (Pimax) and TT performance in normoxia ($R^2 = 0.69$, $p = 0.00$; **Figure 3**) and in hypoxia ($R^2 = 0.67$, $p = 0.00$). No relationship was found between time trial performance (W_{TTmean}) and ventilatory efficiency (V_E/VCO_2 slope) in normoxia ($R^2 = 0.149$, $p = 0.02$; **Figure 4**) and in hypoxia ($R^2 = 0.02$, $p = 0.81$). W_{TTmean} and OUES were significantly related in normoxia ($R^2 = 0.647$, $p = 0.00$; **Figure 5**) and in hypoxia ($R^2 = 0.631$, $p = 0.01$).

DISCUSSION

To the best of our knowledge, this is the first study that investigated the effects of IMT on ventilatory efficiency variables in normoxic and hypoxic conditions. We hypothesized that IMT could improve the ventilatory efficiency response in normoxia and hypoxia. We also hypothesized that improvements in the submaximal cycling performance may be linked to improvements in ventilatory efficiency. The main finding of this study was that IMT improved V_E/VCO_2 slope (−7.95%) in hypoxia and TT performance in both normoxia (+10.17%) and hypoxia (+6.62%) conditions. However, despite this within-group effect no interaction effect was found. Additionally, cycling TT performance was positively related to the OUES in normoxia and hypoxia and to the inspiratory muscle strength (Pimax). These findings partly support (only in OUES) the hypothesis that changes in sport performance after IMT may be linked to changes in ventilatory efficiency.

It is well known that V_E is stimulated in hypoxia mediated by stimulation of the peripheral chemoreceptors (Dempsey and Forster, 1982; Townsend et al., 2002). However, the influence of hypoxic conditions on ventilatory efficiency has not been thoroughly investigated. In the present study, hypoxia at Pre worsened the ventilatory efficiency response in both groups (+18.5% V_E/VCO_2 slope; +9.58% $LEqCO_2$; +14.09% $EqCO_2VT_2$; and −8.64% OUES, respectively; **Table 2**), which is in agreement with the increase in the ventilatory equivalents described at 16% FiO_2 (Ozcelik and Kelestimur, 2004). The increased V_E at altitude, initiated to maintain SaO_2 (Rusko et al., 2004; Burtscher et al., 2006; Faiss et al., 2014), may to some extent explain the deterioration of ventilatory efficiency. In addition, in hypoxia V_E may be increased in excess of what would be required to maintain partial pressure levels of carbon dioxide ($PaCO_2$; Warner and Mitchell, 1990) thus influencing the relationship between V_E and VCO_2.

After the training period, hypoxia did not significantly increase the V_E/VCO_2 slope response in the subjects that performed the IMT [+3.43% (ES = 0.4) vs. +9.48% (ES = 1.3) for the IMTG and CG, respectively]. These changes

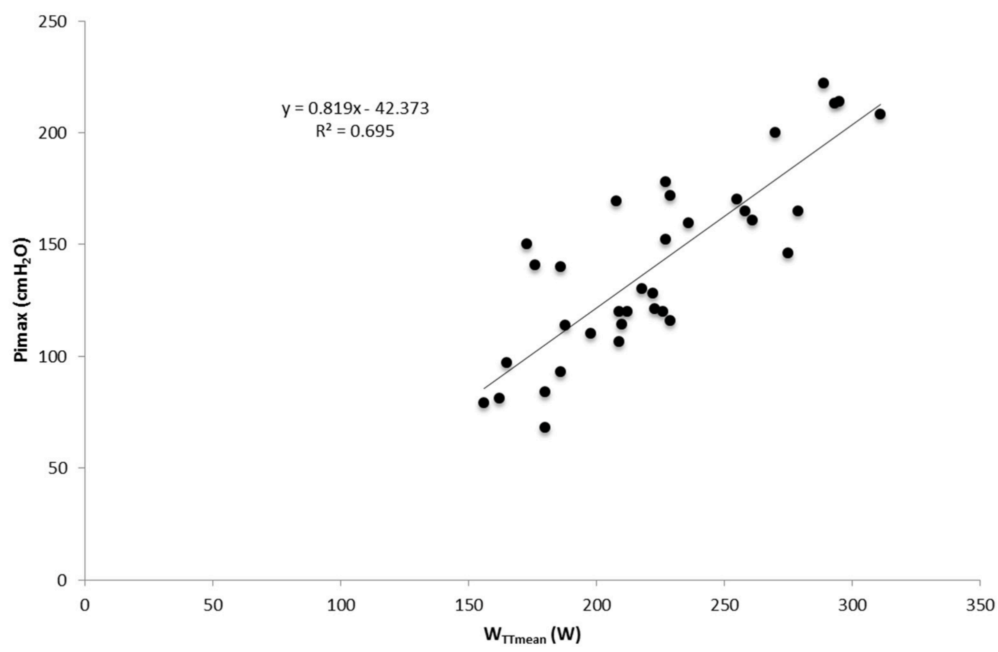

FIGURE 3 | Relationship between inspiratory muscle strength (Pimax) and cycling time trial (TT) performance with data from both groups and both test in normoxia.

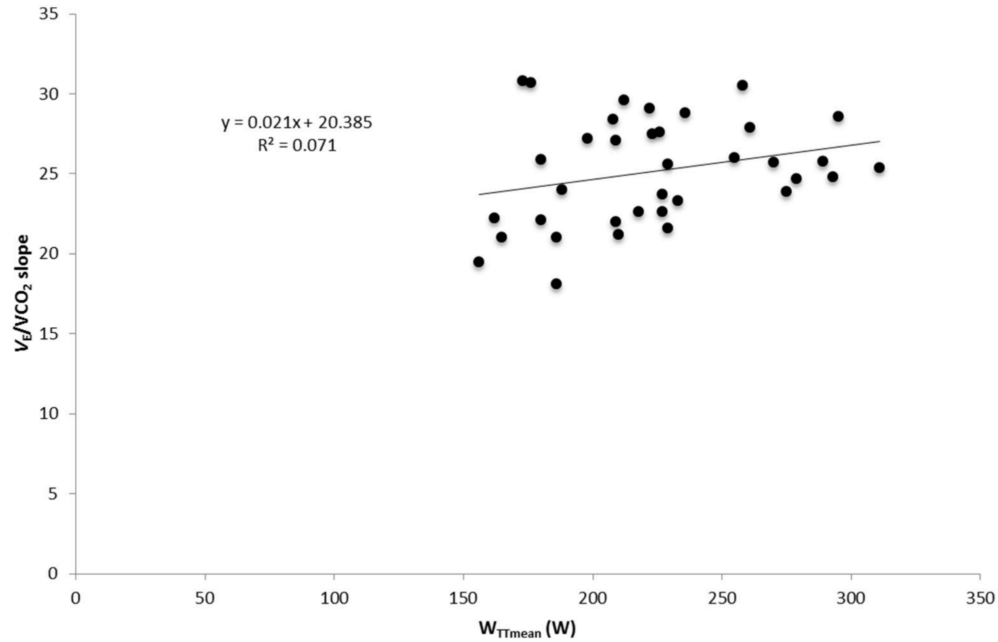

FIGURE 4 | Relationship between cycling time trial (TT) performance and ventilatory efficiency measured as V_E/VCO_2 slope with data from both groups and both test in normoxia.

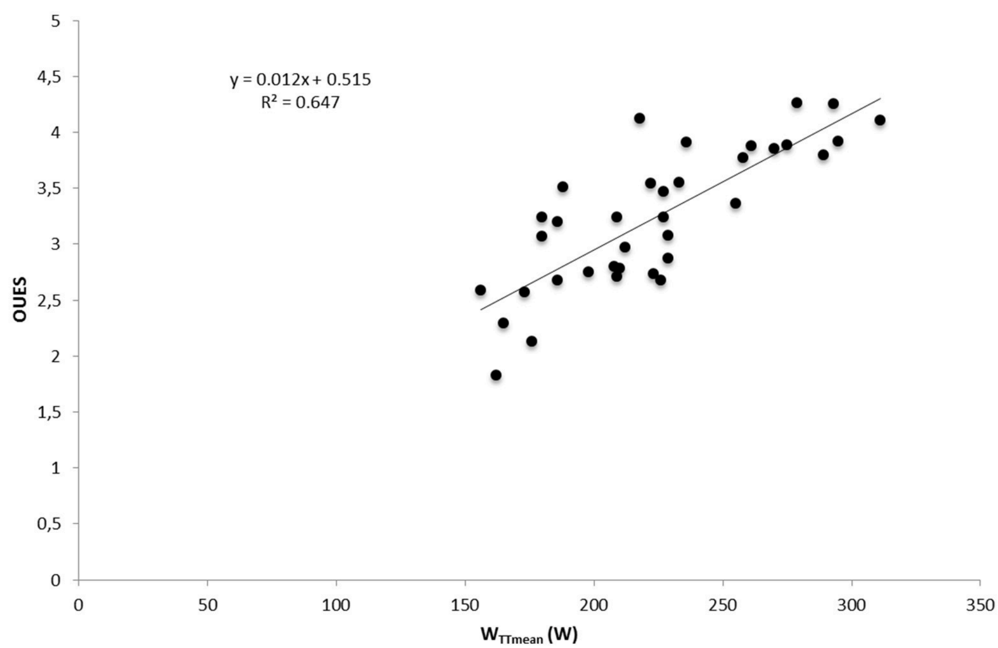

FIGURE 5 | Relationship between cycling time trial (TT) performance and ventilatory efficiency measured as OUES with data from both groups and both test in normoxia.

might be explained by improvements in breathing muscle strength (Pimax) and altered breathing patterns after IMT. Respiratory muscle training has been shown as an effective method to improve A-a gradient and ventilation–perfusion mismatch (Esposito et al., 2010). A better A-a gradient in hypoxia might have reduced the V_E overshoot observed in hypoxia before training (**Table 3**). In support of this, it has been reported that climbers who managed to climb Mt. Everest and K2 without oxygen, are those with a high ventilatory efficiency and "optimized" breathing patterns (Bernardi et al., 2006). Again, it has to be underlined that no interaction effect existed with respect to the altered V_E/VCO_2 slope in hypoxia after the training period. Therefore, the reported training effect contains some uncertainty and further studies with a greater sample size are needed to confirm our conclusions.

With regard to cycling performance, TT performance was reduced significantly in both groups before IMT in hypoxia (**Table 4**). After IMT, only the IMTG improved TT performance in normoxia and hypoxia (**Table 4**). Our results support previous studies showing a positive effect of IMT on sport performance (Volianitis et al., 2001; Romer et al., 2002a,b; Edwards and Walker, 2009). However, the participants of the present study not only improved their performance in normoxia (+11.33%), they also improved their performance in hypoxia (+7.33%) despite a reduction in VO_{2max} (−5.42%; **Table 4**). It could be hypothesized that IMT reduced the oxygen cost of the breathing muscles allowing higher O_2 availability for the locomotor muscles. In addition, it has been suggested that reductions in respiratory effort could lead to greater locomotor muscle recruitment mediated by central nervous system control (Edwards and Walker, 2009). Once more, it should be noted that despite the improvements found in the IMTG no interaction effect was found. Thus, outcomes should be interpreted with some caution.

A further finding of the present investigation was that most of the ventilatory efficiency variables (V_E/VCO_2 slope, $LEqCO_2$ and $EqCO_2VT_2$) were not related to TT performance (**Table 6**; **Figure 5**). This is in contrast to the finding of Sheel (2002) who suggests that improvements in submaximal exercise performance after IMT are related to improvements in ventilatory efficiency. Nonetheless, results of the present investigation show a positive relationship between cycling time trial performance and respiratory muscle strength ($r^2 = 0.695$; **Figure 3**). It could be argued that the increased respiratory muscle strength might reduce the oxygen cost of breathing during submaximal exercise, thus improving oxygen delivery to the working limb muscles. However, further studies are necessary to confirm this hypothesis. In contrast, OUES showed a linear relationship with cycling TT performance ($r^2 = 0.647$; **Figure 5**). Subjects who showed a lower oxygen cost for the same increment in V_E are those who achieved a higher performance in the TT (W_{TTmean}; **Table 6**). However, OUES was not modified by the IMT (**Table 3**) and was not related to Pimax (**Table 6**). Therefore, IMT seems to not play a role in this relationship. It should be mentioned that in contrast to our trained sample, OUES was modified by IMT in patients with heart failure and weakened breathing muscle (Winkelmann et al., 2009).

Regarding the incremental exercise test, VO_{2max} in hypoxia was reduced in both groups (−8.99% IMTG; −11.92% CG). Similar reductions were found previously at this simulated altitude (Lawler et al., 1988; Martin and O'kroy, 1993). Additionally, IMT did not improve VO_{2max} in normoxia and hypoxia (**Table 5**). Our results support previous studies that did not find an effect of IMT on VO_{2max} in normoxia and hypoxia (Downey et al., 2007; Esposito et al., 2010). However, the IMTG improved PPO after IMT in normoxia (+5.62%) and hypoxia

TABLE 2 | Evaluation of ventilatory efficiency variables in normoxia and hypoxia before the experimental period with data from both groups (Mean ± *SD*).

	V_E/VCO_2 slope	$LEqCO_2$	$EqCO_2VT_2$	OUES
Normoxia (*n* = 16)	24.58 ± 2.95	22.63 ± 2.68	23.91 ± 2.34	3.24 ± 0.62
Hypoxia (*n* = 16)	29.15 ± 3.26*	24.8 ± 1.9*	27.28 ± 2.79*	2.96 ± 0.85*
% ΔChange	+18.5%	+9.58%	+14.09%	−8.64%

V_E/VCO_2 slope, Slope of the relationship between VCO_2 and V_E; $LEqCO_2$, lowest equivalent of CO_2 during the incremental test; $EqCO_2VT_2$, equivalent of CO_2 in the second ventilatory threshold; OUES, oxygen uptake efficiency slope; % ΔChange, Percentage of change between measurements.

**t-Test for paired samples ($p < 0.05$).*

TABLE 3 | Comparison between groups in ventilatory efficiency variables in the four experimental conditions.

IMTG	Pre		Post		ANOVA			
	Normoxia	Hypoxia	Normoxia	Hypoxia	Main effect (time)	Main effect (condition)	Main effect (group)	Interaction (condition × group × time)
V_E/VCO_2 slope	23.68 ± 2.94	28.77 ± 2.74# (1.3)	25.6 ± 3.95	26.48 ± 2.77* (0.42)	0.267	0.000	0.476	0.313
$LEqCO_2$	22.51 ± 1.32	24.72 ± 1.45# (1.2)	22.73 ± 1.65	23.98 ± 1.64# (0.83)	0.609	0.000	0.755	0.519
$EqCO_2VT_2$	23.68 ± 2.33	27.26 ± 2.94# (1.3)	24.32 ± 2.92	24.38 ± 2.11* (1.5)	0.022	0.000	0.733	0.233
OUES	3.22 ± 0.75	3.13 ± 0.82	3.31 ± 0.83	2.92 ± 0.71# (0.81)	0.493	0.007	0.994	0.203
CG								
V_E/VCO_2 slope	25.31 ± 2.35	29.81 ± 3.7# (1.7)	25.63 ± 3.93	28.06 ± 3.5*# (1.2* − 1.3#)	–	–	–	–
$LEqCO_2$	22.75 ± 3.59	24.87 ± 2.17# (0.5)	22.62 ± 1.98	24.73 ± 1.58# (1.9)	–	–	–	–
$EqCO_2VT_2$	24.26 ± 2.05	27.15 ± 2.81# (0.8)	24.15 ± 3.11	25.7 ± 2.73* (0.8)	–	–	–	–
OUES	3.36 ± 0.56	2.98 ± 0.91	3.24 ± 0.52	3.01 ± 0.55# (1.0)	–	–	–	–

Data are presented as Mean ± SD and Effect size (ES). ES is showed when a statistical difference was found. V_E/VCO_2 slope, Slope of the relationship between VCO_2 and V_E; $LEqCO_2$, lowest equivalent of CO_2 during the incremental test; $EqCO_2VT_2$, equivalent of CO_2 in the second ventilatory threshold; OUES, oxygen uptake efficiency slope. ANOVA mixed-effects Bonferroni post-hoc test:

**Mixed-effects ANOVA Pre vs. Post in the same condition ($p < 0.05$).*

#Mixed-effects ANOVA Nor vs. Hyp at the same time ($p < 0.05$).

TABLE 4 | Comparison between groups in time trial variables in the four experimental conditions.

IMTG	Pre		Post		ANOVA			
	Normoxia	Hypoxia	Normoxia	Hypoxia	Main effect (time)	Main effect (condition)	Main effect (group)	Interaction (condition × group × time)
W_{TTmean} (W)	217.25 ± 49.07	204.5 ± 49.67# (1.0)	241.87 ± 56.01*(1.9)	219 ± 51.22# * (0.6# −0.6*)	0.026	0.000	0.755	0.611
W_{TTmean} (W/Kg)	3.03 ± 0.4	2.83 ± 0.45# (1.3)	3.35 ± 0.4*(2.4)	3.07 ± 0.44# * (1.8# −1.5*)	0.041	0.000	0.823	0.610
W_{TTmax} (W)	296.25 ± 109.6	282.3 ± 112	319.12 ± 118	289 ± 105.9# (1.8)	0.969	0.000	0.353	0.769
PTF (W)	147.5 ± 22.83	141.25 ± 27.35	156.87 ± 29.51* (1.1)	144.37 ± 27.57# (2.3)	0.466	0.000	0.676	0.164
CG								
W_{TTmean} (W)	221.25 ± 32.5	209.12 ± 37.47# (1.6)	222 ± 35.25	202.62 ± 36.23# (2.9)	–	–	–	–
W_{TTmean} (W/Kg)	3.08 ± 0.39	2.88 ± 0.43# (1.6)	3.11 ± 0.32	2.89 ± 0.32# (2.8)	–	–	–	–
W_{TTmax} (W)	273.28 ± 28.87	258.37 ± 36.89	265 ± 39.84	238.7 ± 37.45# (2.6)	–	–	–	–
PTF (W)	161.87 ± 22.19	146.87 ± 21.86# (1.7)	156.87 ± 22.98	144.75 ± 21.59# (1.0)	–	–	–	–

Data are presented as Mean ± SD and Effect size (ES). ES is showed when a statistical difference was found. W_{TTmax}, Peak power output; W_{TTmean} (W), mean watts; W_{TTmean} (W/Kg), mean watts per kilogram; PTF, pedal torque force. ANOVA mixed-effects Bonferroni post-hoc test.
**Mixed-effects ANOVA Pre vs. Post in the same condition ($p < 0.05$).*
#Mixed-effects ANOVA Nor vs. Hyp at the same time ($p < 0.05$).

TABLE 5 | Measured cardiorespiratory and performance variables at maximal exercise intensity in the four experimental conditions.

IMTG	Pre		Post		ANOVA			
	Normoxia	Hypoxia	Normoxia	Hypoxia	Main effect (time)	Main effect (condition)	Main effect (group)	Interaction (condition × group × time)
VO_{2max} (ml·kg·min^{-1})	47.19 ± 9.45	45.15 ± 7.34	45.86 ± 5.07	43.37 ± 6.88# (0.6)	0.139	0.018	0.973	0.731
PPO (W)	289.37 ± 55.12	274.62 ± 53.28# (0.7)	306.62 ± 58.86* (0.9)	281.5 ± 51.37# (2.0)	0.180	0.000	0.778	0.660
V_{Emax} (l·min^{-1})	141.12 ± 32.24	146.75 ± 34.58	150.37 ± 28.99	143.62 ± 23.46	0.785	0.525	0.919	0.105
V_{Tmax} (l)	3.06 ± 0.79	3.07 ± 0.72	3.04 ± 0.58	3.03 ± 0.65	0.779	0.911	0.628	0.865
BF_{max} (breaths·min^{-1})	57.25 ± 5.54	56.5 ± 7.72	57.12 ± 5.93	56 ± 7.38	0.468	0.711	0.488	0.850
CG								
VO_{2max} (ml·kg·min^{-1})	49 ± 8.37	43.78 ± 7.24	46.51 ± 4.1	42.67 ± 4.06# (2.3)	–	–	–	–
PPO (W)	306.62 ± 41.86	285.5 ± 43.1# (2.1)	307.75 ± 47.67	280.25 ± 44.65# (1.9)	–	–	–	–
V_{Emax} (l·min^{-1})	143.5 ± 35.97	148.5 ± 42.57	135.87 ± 40.92	147.12 ± 43.6	–	–	–	–
VT_{max} (l)	2.87 ± 0.75	2.93 ± 0.77	2.88 ± 0.8	2.87 ± 0.82	–	–	–	–
BF_{max} (breaths·min^{-1})	59.25 ± 10.87	62 ± 9.81	57.75 ± 14	59.12 ± 11.24	–	–	–	–

Data are presented as Mean ± SD and Effect size (ES). ES is showed when a statistical difference was found. VO_{2max}, Maximum oxygen uptake; PPO, peak power output; V_{Emax}, maximum ventilation; VT_{max}, maximum tidal volume; BF_{max}, maximum breathing frequency. ANOVA mixed-effects Bonferroni post hoc test:
**Mixed-effects ANOVA Pre vs. Post in the same condition ($p < 0.05$).*
#Mixed-effects ANOVA Nor vs. Hyp at the same time ($p < 0.05$).

(+2.51%) which is in contrast to previous studies that reported only a slight influence of IMT on PPO (Sheel, 2002; Illi et al., 2012). Moreover, except for OUES, the ventilatory efficiency variables were not correlated with performance variables of the incremental test (**Table 6**). Similar to the time trial outcome, OUES showed a strong correlation with VO_{2max} in normoxia and hypoxia ($r = 0.89$ and $r = 0.82$; respectively) and with PPO ($r = 0.91$ and $r = 0.88$; respectively). With respect to this finding, there are contrasting results reported in the literature. There are studies reporting a correlation between VO_{2max} and OUES (Baba et al., 1999a,b,c; Hollenberg and Tager, 2000) and others that did not find a correlation between these two parameters or only a weak correlation (Brown, 2010; Brown et al., 2013). Further research is necessary on the influence of IMT on ventilatory efficiency parameters in hypoxia.

Some limitations have to be addressed. First, the sample size was large enough to detect changes in Pimax and performance within the intervention group but might have been too low to detect group differences. Second, we do not have exact data on the amount of training and competitions completed by the subjects apart from the inspiratory training load. However, participants were advised not to change their usual training habits during the experimental period. All participants were enrolled in the same practical courses, and all reported to only

TABLE 6 | Correlation analysis between performance variables and ventilatory efficiency variables after experimental protocol with data from both groups.

	Pearson-*r*				
	W_{TTmean} (W)	VO_{2max} ($ml \cdot kg \cdot min^{-1}$)	V_{Emax} ($l \cdot min^{-1}$)	Pi_{max} (cmH_2O)	PPO (W)
NORMOXIA					
$Pimax_{(cmH_2O)}$	0.607*	0.503*	0.859*	1	0.623*
V_E/VCO_2 slope	0.126	0.153	0.278	0.361	0.036
$LEqCO_2$	0.011	0.083	0.288	0.274	−0.064
$EqCO_2VT_2$	−0.1	0.026	0.062	0.196	−0.220
OUES	0.89*	0.683*	0.669*	0.454	0.913*
HYPOXIA					
Pimax (cmH_2O)	0.599*	0.477	0.545*	1	0.587*
V_E/VCO_2 slope	0.060	0.045	0.145	0.304	0.029
$LEqCO_2$	−0.250	−0.084	−0.029	0.069	−0.283
$EqCO_2VT_2$	−0.105	−0.016	−0.019	0.131	−0.166
OUES	0.828*	0.664*	0.79*	0.408	0.885*

Pimax, Peak inspiratory pressure; V_E/VCO_2 slope, Slope of the relationship between VCO_2 and V_E; $LEqCO_2$, lowest equivalent of CO_2 during the incremental test; $EqCO_2VT_2$, equivalent of CO_2 in the second ventilatory threshold; OUES, oxygen uptake efficiency slope; VO_{2max}, Maximum oxygen uptake; PPO, peak power output; VEmax, maximum ventilation; WTTmean, mean watts.

**Significant correlation ($p < 0.05$).*

have limited time for sports outside of the university setting. In addition, it was reported that conventional training (no specific breathing training) does not improve breathing muscle strength (Illi et al., 2012). Therefore, we can assume that they completed approximately the same training apart from the IMT and this did not influence our results. Third, we did not measure the oxygen saturation (SaO_2) during the hypoxia trials and this may have contributed important information. Lastly, we did not control for a possible placebo effect. However, as it is stated in the methods section, a large placebo effect is not expected. Therefore, we are confident that our conclusions were not affected.

CONCLUSIONS

Even though sample size might have been too low to show an interaction effect, the results of the present study suggest a possible positive effect of IMT on cycling time trial performance in both normoxic and hypoxic conditions. Additionally, this study shows that hypoxia has a negative effect on the ventilatory efficiency and that IMT may reduce this effect. Finally, the data suggest that except or OUES, ventilatory efficiency measures seem not to affect cycling time trial performance. These findings may have relevance for athletes planning to complete a high altitude training camp or for athletes competing at high altitude. IMT before a competition at altitude might be a successful method to improve performance.

ETHICS STATEMENT

This study was carried out in accordance with the recommendations of Ethics Committee of the University of Innsbruck with written informed consent from all subjects. All subjects gave written informed consent in accordance with the Declaration of Helsinki. The protocol was approved by the Ethics Committee of the University of Innsbruck.

AUTHOR CONTRIBUTIONS

Conception and design of the experiments: ES, AS, and JNO; Pre-testing, experimental preparation, data collection, and analysis: ES and HG. The first version of the manuscript was written by ES, HG, MB, JNO, and AS. All co-authors read and approved the final version of the manuscript.

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REFERENCES

Arena, R., Myers, J., and Guazzi, M. (2008). The clinical and research applications of aerobic capacity and ventilatory efficiency in heart failure: an evidence-based review. *Heart Fail. Rev.* 13, 245–269. doi: 10.1007/s10741-007-9067-5

Baba, R., Kubo, N., Morotome, Y., and Iwagaki, S. (1999a). Reproducibility of the oxygen uptake efficiency slope in normal healthy subjects. *J. Sports Med. Phys. Fitness* 39, 202–206.

Baba, R., Nagashima, M., Goto, M., Nagano, Y., Yokota, M., Tauchi, N., et al. (1996). Oxygen intake efficiency slope: a new index of cardiorespiratory functional reserve derived from the relationship between oxygen consumption and minute ventilation during incremental exercise. *J. Am. Coll. Cardiol.* 59, 55–62.

Baba, R., Nagashima, M., Nagano, Y., Ikoma, M., and Nishibata, K. (1999b). Role of the oxygen uptake efficiency slope in evaluating exercise tolerance. *Arch. Dis. Child.* 81, 73–75. doi: 10.1136/adc.81.1.73

Baba, R., Tsuyuki, K., Kimura, Y., Ninomiya, K., Aihara, M., Ebine, K., et al. (1999c). Oxygen uptake efficiency slope as a useful measure of cardiorespiratory functional reserve in adult cardiac patients. *Eur. J. Appl. Physiol. Occup. Physiol.* 80, 397–401. doi: 10.1007/s004210050610

Babcock, M. A., Johnson, B. D., Pegelow, D. F., Suman, O. E., Griffin, D., and Dempsey, J. A. (1995). Hypoxic effects on exercise-induced diaphragmatic fatigue in normal healthy humans. *J. Appl. Physiol.* 78, 82–92.

Bernardi, L., Schneider, A., Pomidori, L., Paolucci, E., and Cogo, A. (2006). Hypoxic ventilatory response in successful extreme altitude climbers. *Eur. Respir. J.* 27, 165–171. doi: 10.1183/09031936.06.00015805

Brown, S. J. (2010). Cardio-respiratory system efficiency in trained endurance cyclists. *Med. Sportiva* 14, 176–181. doi: 10.2478/v10036-010-0028-z

Brown, S. J., Raman, A., Schlader, Z., and Stannard, S. R. (2013). Ventilatory efficiency in juvenile elite cyclists. *J. Sci. Med. Sport* 16, 266–270. doi: 10.1016/j.jsams.2012.06.010

Burtscher, M., Faulhaber, M., Flatz, M., Likar, R., and Nachbauer, W. (2006). Effects of short-term acclimatization to altitude (3200 m) on aerobic and anaerobic exercise performance. *Int. J. Sports Med.* 27, 629–635. doi: 10.1055/s-2005-872823

Dempsey, J. A., and Forster, H. V. (1982). Mediation of ventilatory adaptations. *Physiol. Rev.* 62, 262–346.

Downey, A. E., Chenoweth, L. M., Townsend, D. K., Ranum, J. D., Ferguson, C. S., and Harms, C. A. (2007). Effects of inspiratory muscle training on exercise responses in normoxia and hypoxia. *Respir. Physiol. Neurobiol.* 156, 137–146. doi: 10.1016/j.resp.2006.08.006

Edwards, A. M., and Walker, R. E. (2009). Inspiratory muscle training and endurance: a central metabolic control perspective. *Int. J. Sports Physiol. Perform.* 4, 122–128. doi: 10.1123/ijspp.4.1.122

Esposito, F., Limonta, E., Alberti, G., Veicsteinas, A., and Ferretti, G. (2010). Effect of respiratory muscle training on maximum power in normoxia and hypoxia. *Respir. Physiol. Neurobiol.* 170, 268–272. doi: 10.1016/j.resp.2010.02.004

Faiss, R., Von Orelli, C., Deriaz, O., and Millet, G. P. (2014). Responses to exercise in normobaric hypoxia: comparison of elite and recreational ski mountaineers. *Int. J. Sports Physiol. Perform*. 9, 978–984. doi: 10.1123/ijspp.2013-0524

Harms, C. A., Babcock, M. A., McClaran, S. R., Pegelow, D. F., Nickele, G. A., Nelson, W. B., et al. (1997). Respiratory muscle work compromises leg blood flow during maximal exercise. *J. Appl. Physiol.* 82, 1573–1583.

Hollenberg, M., and Tager, I. B. (2000). Oxygen uptake efficiency slope: an index of exercise performance and cardiopulmonary reserve requiring only submaximal exercise. *J. Am. Coll. Cardiol.* 36, 194–201. doi: 10.1016/S0735-1097(00)00691-4

Howley, E. T., Bassett, D. R., and Welch, H. G. (1995). Criteria for maximal oxygen uptake: review and commentary. *Med. Sci. Sports Exerc.* 27, 1292–1301. doi: 10.1249/00005768-199509000-00009

Illi, S. K., Held, U., Frank, I., and Spengler, C. M. (2012). Effect of respiratory muscle training on exercise performance in healthy individuals. *Sports Med.* 42, 707–724. doi: 10.1007/BF03262290

Ingle, L., Goode, K., Carroll, S., Sloan, R., Boyes, C., Cleland, J. G. F., et al. (2007). Prognostic value of the V_E/VCO_2 slope calculated from different time intervals in patients with suspected heart failure. *Int. J. Cardiol.* 118, 350–355. doi: 10.1016/j.ijcard.2006.07.105

Laveneziana, P., Agostoni, P., Mignatti, A., Mushtaq, S., Colombo, P., Sims, D., et al. (2010). Effect of acute β-blocker withholding on ventilatory efficiency in patients with advanced chronic heart failure. *J. Card. Fail.* 16, 548–555. doi: 10.1016/j.cardfail.2010.02.006

Lawler, J., Powers, S. K., and Thompson, D. (1988). Linear relationship between VO_{2max} and VO_{2max} decrement during exposure to acute hypoxia. *J. Appl. Physiol.* 64, 1486–1492.

Lucía, A., Hoyos, J., Pérez, M., and Chicharro, J. L. (2000). Heart rate and performance parameters in elite cyclists: a longitudinal study. *Med. Sci. Sports Exerc.* 32, 1777–1782. doi: 10.1097/00005768-200010000-00018

Magrì, D., Limongelli, G., Re, F., Agostoni, P., Zachara, E., Correale, M., et al. (2016). Cardiopulmonary exercise test and sudden cardiac death risk in hypertrophic cardiomyopathy. *Heart* 102, 602–609. doi: 10.1136/heartjnl-2015-308453

Martin, D., and O'kroy, J. (1993). Effects of acute hypoxia on the VO2 max of trained and untrained subjects. *J. Sports Sci.* 11, 37–42. doi: 10.1080/02640419308729961

Mourot, L., Perrey, S., Tordi, N., and Rouillon, J. D. (2004). Evaluation of fitness level by the oxygen uptake efficiency slope after a short-term intermittent endurance training. *Int. J. Sports Med.* 25, 85–91. doi: 10.1055/s-2004-819943

Ozcelik, O., and Kelestimur, H. (2004). Effects of acute hypoxia on the estimation of lactate threshold from ventilatory gas exchange indices during an incremental exercise test. *Physiol. Res.* 53, 653–659.

Romer, L. M., McConnell, A. K., and Jones, D. A. (2002a). Effects of inspiratory muscle training on time-trial performance in trained cyclists. *J. Sports Sci.* 20, 547–590. doi: 10.1080/026404102760000053

Romer, L. M., McConnell, A. K., and Jones, D. A. (2002b). Inspiratory muscle fatigue in trained cyclists: effects of inspiratory muscle training. *Med. Sci. Sports Exerc.* 34, 785–792. doi: 10.1097/00005768-200205000-00010

Roussos, C. (1985). Function and fatigue of respiratory muscles. *Chest* 88, 124S–132S. doi: 10.1378/chest.88.2_Supplement.124S

Rusko, H., Tikkanen, H., and Peltonen, J. (2004). Altitude and endurance training. *J. Sports Sci.* 22, 928–945. doi: 10.1080/02640410400005933

Salazar-Martínez, E., Terrados, N., Burtscher, M., Santalla, A., and Orellana, J. N. (2016). Ventilatory efficiency and breathing pattern in world-class cyclists: a three-year observational study. *Respir. Physiol. Neurobiol.* 229, 17–23. doi: 10.1016/j.resp.2016.04.001

Sheel, A. W. (2002). Respiratory muscle training in healthy individuals. *Sports Med.* 32, 567–581. doi: 10.2165/00007256-200232090-00003

Sun, X.-G., Hansen, J. E., Garatachea, N., Storer, T. W., and Wasserman, K. (2002). Ventilatory efficiency during exercise in healthy subjects. *Am. J. Respir. Crit. Care Med.* 166, 1443–1448. doi: 10.1164/rccm.2202033

Townsend, N. E., Gore, C. J., Hahn, A. G., McKenna, M. J., Aughey, R. J., Clark, S. A., et al. (2002). Living high-training low increases hypoxic ventilatory response of well-trained endurance athletes. *J. Appl. Physiol.* 93, 1498–1505. doi: 10.1152/japplphysiol.00381.2002

Volianitis, S., McConnell, A. K., Koutedakis, Y., McNaughton, L. R., Backx, K., and Jones, D. A. (2001). Inspiratory muscle training improves rowing performance. *Med. Sci. Sports Exerc.* 33, 803–809. doi: 10.1097/00005768-200105000-00020

Warner, M. M., and Mitchell, G. S. (1990). Ventilatory responses to hyperkalemia and exercise in normoxic and hypoxic goats. *Respir. Physiol.* 82, 239–249. doi: 10.1016/0034-5687(90)90038-Z

Wells, G. D., Plyley, M., Thomas, S., Goodman, L., and Duffin, J. (2005). Effects of concurrent inspiratory and expiratory muscle training on respiratory and exercise performance in competitive swimmers. *Eur. J. Appl. Physiol.* 94, 527–540. doi: 10.1007/s00421-005-1375-7

Winkelmann, E. R., Chiappa, G. R., Lima, C. O. C., Viecili, P. R. N., Stein, R., and Ribeiro, J. P. (2009). Addition of inspiratory muscle training to aerobic training improves cardiorespiratory responses to exercise in patients with heart failure and inspiratory muscle weakness. *Am. Heart* 158, 768.e1–e7. doi: 10.1016/j.ahj.2009.09.005

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Influence of high-intensity interval training on ventilatory efficiency in trained athletes

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Abstract:	The aim of this study was to investigate the effects of 3-week high-intensity interval training (HIIT) on ventilatory efficiency (VE/VCO2 slope) in endurance athletes. Sixteen male well-trained (67.72 ml·kg·min-1) sport students participated in this study. Each participant performed an incremental exercise test with gas analysis (i.e. VE, VO2) and a 400 m running field test (T400m) before and after the 3-week intervention period. HIIT group (HIITG) performed 11 HIIT sessions consisting of four 4-min interval bouts at an exercise intensity of 90–95% of the VO2max, separated by 4-min active recovery periods (work/rest ratio = 1:1). No significant differences were found in the parameters studied. Ventilatory efficiency (up to VT2 and up to exhaustion) did not show any change in HIITG after training intervention (ES=0.24 HIITG and ES=0.21 CG). No significant changes were observed on ventilation (VEmax), but HIITG showed an increase on VEmax after training (ES=0.38). VO2max and T400m did not show a significant improvement after the training period (no interaction time x group, p<0.05) (ES=0.43 and ES=0.75 respectively). These results do not support the hypothesis that 3 weeks of HIIT could modify the ventilatory efficiency response in well-trained athletes. Furthermore, they show the lack of relationship between ventilatory efficiency and sport performance.

Influence of high-intensity interval training on ventilatory efficiency in trained athletes

ABSTRACT

The aim of this study was to investigate the effects of 3-week high-intensity interval training (HIIT) on ventilatory efficiency (V_E/VCO_2 slope) in endurance athletes. Sixteen male well-trained (67.72 $ml \cdot kg \cdot min^{-1}$) sport students participated in this study. Each participant performed an incremental exercise test with gas analysis (i.e. V_E, VO_2) and a 400 m running field test (T400m) before and after the 3-week intervention period. HIIT group (HIITG) performed 11 HIIT sessions consisting of four 4-min interval bouts at an exercise intensity of 90–95% of the VO_{2max}, separated by 4-min active recovery periods (work/rest ratio = 1:1). No significant differences were found in the parameters studied. Ventilatory efficiency (up to VT_2 and up to exhaustion) did not show any change in HIITG after training intervention (ES=0.24 HIITG and ES=0.21 CG). No significant changes were observed on ventilation (V_{Emax}), but HIITG showed an increase on V_{Emax} after training (ES=0.38). VO_{2max} and T400m did not show a significant improvement after the training period (no interaction time x group, $p<0.05$) (ES=0.43 and ES=0.75 respectively). These results do not support the hypothesis that 3 weeks of HIIT could modify the ventilatory efficiency response in well-trained athletes. Furthermore, they show the lack of relationship between ventilatory efficiency and sport performance.

Key words: V_E/VCO_2 slope / HIIT / ventilation / chemosensitivity / training

1. Introduction

High-Intensity Interval Training (HIIT) is defined as either repeated short (<45 s) to long (2-4 min) bouts of rather high-but not maximal intensity exercise, or short (<10 s, repeated-sprint sequences (RSS)) or long (>20-30 s, sprint interval session (SIT)) all-out sprints interspersed with recovery periods [4]. HIIT has increased its popularity during recent years as an effective method to improve exercise performance [2]. The mechanisms proposed which could explain the improvements in sport performance after HIIT programs are: increments in VO_{2max} [2,16], changes in plasma volume [26], changes in hormonal and metabolic response [30] or increments in skeletal muscle oxidative capacity [9,13]. However, the influence of HIIT on ventilatory parameters is still discussed controversially. Previous studies only analysed ventilation (V_E) response after HIIT [8,19]. V_E determined at a fixed submaximal speed did not change after 12-week of HIIT in athletes who completed the training intervention period [19]. Maximum ventilation (V_{Emax}) and maximal breathing frequency (f_{Rmax}) did not change after 3-week of HIIT neither in normoxia or hypoxia in basketball players [8]. However, the influence of HIIT on ventilatory efficiency has been only studied in patients. No changes were observed on ventilatory efficiency after a HIIT program in chronic heart failure patients [24] and coronary heart disease subjects [6]. The limited studies in this area have reported that ventilatory efficiency generally improves after training, although this is not an entirely consistent finding [24]. In athletes, no changes on ventilatory efficiency (measured as V_E/VCO_2 slope) were found after 16-week of non-controlled training in juvenile cyclists [3]. Similar results were found in world-class cyclists, ventilatory efficiency did not change after 3 competitive seasons [28]. However, from our knowledge there are not studies which evaluated the influence of a HIIT program on ventilatory efficiency in well-trained athletes. In order to better clarify the influence of exercise on ventilatory efficiency in athletes, the present study aimed to investigate the effects of 3-week of HIIT on V_E/VCO_2 slope in athletes. We hypothesized that 3-week of HIIT could promote changes on the ventilatory efficiency response of well-trained athletes due to the inter-individual adjustment in breathing pattern described at high exercise intensities [15] and the high ventilatory requirements observed during HIIT sessions [31] .

2. Materials and methods

2.1. Participants

Sixteen male well-trained sport students participated in the study (Table 1). Participants underwent a routine pre-participation screening prior to the baseline testing. Inclusion criteria were: a) experienced athletes (≥ 3 years of practice) and b) subjects with a VO_{2max} higher than 60 $ml \cdot kg \cdot min^{-1}$. Exclusion criteria were all types of acute and chronic diseases, taking medication on a regular basis or smoking. The study was carried out according to the Declaration of Helsinki and was approved by the Institutional Review Board of the Department of Sport Science (University Innsbruck). All participants gave written informed consent to participate in the study. Some of these participants were included previously in the sample size of other study [21]. However, we carried out a new analysis about ventilatory efficiency and performance variables never published before.

2.2. Design

The study was designed as a randomized controlled training study including HIIT and control group (HIITG and CG, respectively) and two measurement times (pre-training *vs* post-training). Baseline measurements included a laboratory incremental treadmill test and a 400m running field test. After baseline measurements, the participants were randomly assigned, stratified by VO_{2max} either to the HIITG or the CG. The HIITG started the 3-week running HIIT program whereas CG maintained their usual training during this period. In particular, they were advised not to include additional high-intensity training. The training data for the HIITG and the CG were recorded in a training log book and the total endurance training loads were determined according to [10] as perceived exertion × endurance training session time (Table 2).

2.3. Methodology

2.3.1. Treadmill and 400m running test

The treadmill protocol was performed according to Burtscher, Gatterer, Faulhaber, et al. [5]. Gas analysis was performed using an open spirometric system (Oxycon Mobile, Care Fusion, Würzburg, Germany) which was calibrated before each measurement. Cardio-respiratory parameters (i.e. V_E, VO_2, VCO_2, HR) were recorded breath by breath during the ergospirometry. A test was considered maximal when three of the four criteria proposed by [7] were fulfilled. Additionally, participants carried out a 400m running filed test. The time to complete the 400m was selected as a performance variable (T400m). Athletes were encouraged to achieve their best performance. Post-training measurements were the same as for the baseline condition (relative humidity: 45-65%; temperature: 24-25°C) and were conducted 5 ± 2 days after the last HIIT session.

2.3.2. HIIT program

The HIITG performed 11 HIIT sessions during the 3-week HIIT period. Each HIIT session consisted of four 4-min interval-running bouts at an exercise intensity of 90–95% of the VO_{2max}, separated by 4-min active recovery periods (work/rest ratio = 1:1). During the first week, athletes completed three HIIT sessions. In the following two weeks athletes performed four HIIT sessions each week. Training intensity was controlled by continuous heart rate (HR) monitoring (Polar, Kempele, Finland) and was equivalent to the HR at 90-95% of their VO_{2max} [11]. The rating of perceived exertion (RPE) was determined according to the Borg scale (6–20) [1].

2.3.3. Ventilatory efficiency

The V_E/VCO_2 slope was calculated from the slope of the relationship between VCO_2 and V_E during each incremental exercise test. V_E/VCO_2 slope was calculated from the beginning of the test until the second ventilatory threshold (VT_2) and up to exhaustion. VT_2 was identified by an increase in the ventilatory equivalent of CO_2 ($EqCO_2$) and a decrease in end tidal partial pressure of carbon dioxide ($PETCO_2$) [20].

2.4. Statistical analysis

Data are expressed as mean ± SD and with Cohen's d effect size (ES) for each variable. Normal distribution of data was tested by the Kolmogorov–Smirnov test. A two-way analysis (group × time) of variance (ANOVA) with repeated measurements was used to verify between-group changes. In addition, paired student's t tests were carried out to evaluate within-group effects. The relationships between ventilatory efficiency (V_E/VCO_2 slope) and performance variables (VO_{2max} and T400m) were assessed by regression analyses. The level of significance was set at $p<0.05$ for each statistical analysis. An ES of <0.2 was considered small, 0.5 medium and >0.8 large. Because the influence of HIIT on ventilatory efficiency has never not been studied in athletes, power calculation (G*Power 3.1.7) was based on the present results (post-hoc test).

3. Results

No significant changes were found in the parameters studied (Table 3). Statistical power for the chosen sample size of 16 (8 in the HIITG and 8 in the CG) was 95 %; alpha = 0.05 based on V_E/VCO_2 slope post-hoc data considering no changes on ventilatory efficiency after HIIT. Ventilatory efficiency (up to VT_2 and up to exhaustion) did not show any change in HIITG after training intervention ($p>0.05$; ES=0.24 HIITG and ES=0.21 CG). No significant changes were observed in V_{Emax} (p>005; ES=0.38). VO_{2max} and T400m did not show a significant improvement after the training period (no interaction time x group, $p>0.05$) (ES=0.43 and ES=0.75 in HIITG respectively). No relationship was found between ventilatory efficiency and VO_{2max} ($r^2=0.210$) and sport performance (T400m; $r^2=0.057$)

4. Discussion

To the best of our knowledge, this is the first study investigating the influence of HIIT on ventilatory efficiency in well-trained athletes. We hypothesized that 3 weeks of HIIT could promote changes on ventilatory efficiency in well-trained athletes. However, the main finding of this study was that 3 weeks of HIIT did not modify the ventilatory efficiency response of well-trained athletes. Our results also suggest the lack of relationship between ventilatory efficiency and sport performance. These results agree with previous studies [3,27,28].

Conditions where the CO_2 production is elevated, as high intensity exercise, seem to play an essential role in the ventilatory control [22]. Regular exercise training has been suggested as an effective stimulus which could modify CO_2 chemosensitivity in athletes [18,23,25]. However, uncontrolled training did not show any effect in the ventilatory efficiency response of cyclists [3]. [28]. But the main difference of these studies with ours is that we controlled the training load completed by the participants. However, controlled training did not influenced ventilatory efficiency response in our subjects (Table 3). We measured ventilatory efficiency not only up to VT_2, but also up to exhaustion for investigating its response after the quimio-compensation point where the highest rates of CO_2 production are developed. We investigated whether this stimulus has any effect on V_E *vs* VCO_2 relationship. However, HIIT did not modify ventilatory efficiency response up to VT_2 neither up to exhaustion (Table 3). In the same way, maximum VCO_2 production did not show a significant change after the training period. Our results might suggest that V_E *vs* VCO_2 relationship is hard to modify with training, even with training sessions developed at high CO_2 production ratios (~5.5 $l{\cdot}min^{-1}$) and with a high ventilatory requirements (~185 $l{\cdot}min^{-1}$). According to that, the efficiency of the CO_2 elimination during exercise might move within very tight ranges, could be set by the system and might be hardly modified through training [28]. Increments in CO_2 production may be linked to proportional increment in V_E in spite of a possible inter-individual adjustment in breathing patter at high exercise intensities [15]. This is an important finding, as it might indicate that ventilatory efficiency could be an inborn characteristic which response relative stable in healthy athletes independently of the improvements reported in sport performance.

In addition to this finding, these results suggest that there is not relationship between ventilatory efficiency (V_E/VCO_2 slope) and sport performance (Figure 1, 2). Our results agree with previous evidence reported [3,27,28], and could indicate that the ability to eliminate CO_2 during exercise (greater ventilatory efficiency) does not influence the capacity for achieving a high sport performance. Although the mechanisms which could explain the sport performance improvements after HIIT have been already studied, from our knowledge this is the first time that it has been reported the influence of HIIT on ventilatory efficiency in well-trained athletes.

With reference to V_{Emax}, it increased slightly after the 3 weeks training period in the HIITG (+ 1.19%; ES=0.38) despite the high baseline level reported (181.8±17.8 ml·min^{-1}). Although, there was not a significant difference between pre and post-test (p>0.05). The inter-individual adjustment in breathing pattern described at high exercise intensities [15] and the high ventilatory requirements reported during HIIT sessions [31] could explain these small changes. However, further studies are necessary in order to better clarify this point.

3 weeks of HIIT improved VO_{2max} (+ 2.01 %; Table 3). However, no significant differences were found in the HIITG after the intervention period (p>0.05). Menz, Strobl, Faulhaber, et al. [21] and Helgerud, Høydal, Wang, et al. [17] found greater improvements after three and four weeks of HIIT using the same training protocol (4 x 4 min-intervals), + 3.5% and + 7.2%, respectively. In contrasts to our study, they used a larger sample. In addition, the baseline levels described were lower in comparison with our subjects (55.5 ± 7.4 and 63.6 ± 7.5 vs 68.4 ± 2.7 ml·kg·min^{-1}).

HIIT showed a positive effect in running performance (T400m) (medium-large ES, Table 3). A reduction of -2.18% was found in the total time to complete 400m in HIITG. Our results clearly agree with previous studies which reported the positive effects of HIIT in running performance [12,14,29]. However, once more, it should be noted that despite the improvements found in the HIITG, no significant changes and no interaction effect were found. Thus, the outcomes should be interpreted with caution.

A few limitations must be addressed. Firstly, it could be possible that the intervention period (3 weeks) had not been long enough to promote changes in ventilatory response. Lastly, the small sample size may not have been large enough to detect an interaction effect in sport performance variables.

5. Conclusions

Present data suggests that high-intensity interval training might be beneficial for improving performance in only 3 weeks. This finding may have relevance for athletes planning to complete in a short time period. However, these results do not support the hypothesis that 3 weeks of HIIT could modify the ventilatory efficiency of well-trained athletes. Furthermore, they show a lack of relationship between ventilatory efficiency and sport performance. Thus, this study strengthens the previous evidence reported which supports the idea that ventilatory efficiency could be an innate characteristic which reacts in a tiny range independently of sport performance. This data could help to carry out further analysis of ventilatory efficiency in athletes.

References

1. Borg GA. Psychophysical bases of perceived exertion. Med sci sports exerc 1982; 14: 377-381
2. Breil FA, Weber SN, Koller S, Hoppeler H, Vogt M. Block training periodization in alpine skiing: effects of 11-day HIT on VO2max and performance. Eur J Appl Physiol 2010; 109: 1077-1086
3. Brown SJ, Raman A, Schlader Z, Stannard SR. Ventilatory efficiency in juvenile elite cyclists. J Sci Med. Sport 2013; 16: 266-270
4. Buchheit M, Laursen PB. High-intensity interval training, solutions to the programming puzzle. Sports Med 2013; 43: 313-338
5. Burtscher M, Gatterer H, Faulhaber M, Gerstgrasser W, Schenk K. Effects of intermittent hypoxia on running economy. Int J Sports Med 2010; 31: 644-650
6. Cardozo GG, Oliveira RB, Farinatti PT. Effects of high intensity interval versus moderate continuous training on markers of ventilatory and cardiac efficiency in coronary heart disease patients. ScientificWorldJournal 2015; 2015:
7. Cunha F, Midgley A, Monteiro W, Farinatti P. Influence of cardiopulmonary exercise testing protocol and resting VO2 assessment on% HRmax,% HRR,% VO2max and% VO2R relationships. Int J Sports Med 2010; 31: 319-326
8. Czuba M, Zając A, Maszczyk A, Roczniok R, Poprzęcki S, Garbaciak W, Zając T. The effects of high intensity interval training in normobaric hypoxia on aerobic capacity in basketball players. J Hum Kinet 2013; 39: 103-114
9. Egan B, Carson BP, Garcia□Roves PM, Chibalin AV, Sarsfield FM, Barron N, McCaffrey N, Moyna NM, Zierath JR, O'Gorman DJ. Exercise intensity□dependent regulation of peroxisome proliferator□activated receptor γ coactivator□1α mRNA abundance is associated with differential activation of upstream signalling kinases in human skeletal muscle. J Physiol 2010; 588: 1779-1790
10. Foster C, Florhaug JA, Franklin J, Gottschall L, Hrovatin LA, Parker S, Doleshal P, Dodge C. A new approach to monitoring exercise training. J Strength Cond Res 2001; 15: 109-115
11. García-Pallarés J.; Morán-Navarro R. Propuesta metodológica para el entrenamiento de la resistencia cardiorrespiratoria. J Sport Health Sci Reser 2012; 4: 119-136
12. García-Pinillos F, Cámara-Pérez JC, Soto-Hermoso VM, Latorre-Román PÁ. A High Intensity Interval Training (HIIT)-Based Running Plan Improves Athletic Performance by Improving Muscle Power. J Strength Cond Res 2017; 31: 146-153

13. Gibala MJ, Little JP, Van Essen M, Wilkin GP, Burgomaster KA, Safdar A, Raha S, Tarnopolsky MA. Short term sprint interval versus traditional endurance training: similar initial adaptations in human skeletal muscle and exercise performance. J Physiol 2006; 575: 901-911
14. Gojanovic B, Shultz R, Feihl F, Matheson G. Overspeed HIIT in Lower-Body Positive Pressure Treadmill Improves Running Performance. Med Sci Sports Exerc 2015; 47: 2571-2578
15. Gravier G, Delliaux S, Delpierre S, Guieu R, Jammes Y. Inter-individual differences in breathing pattern at high levels of incremental cycling exercise in healthy subjects. Respir Physiol Neurobiol 2013; 189: 59-66
16. Helgerud J, Hoydal K, Wang E, Karlsen T, Berg P, Bjerkaas M, Simonsen T, Helgesen C, Hjorth N, Bach R. Aerobic High-Intensity Intervals Improve VO2max More Than Moderate Training. Med Sci Sports Exerc 2007; 39: 665
17. Helgerud J, Høydal K, Wang E, Karlsen T, Berg P, Bjerkaas M, Simonsen T, Helgesen C, Hjorth N, Bach R. Aerobic high-intensity intervals improve VO2max more than moderate training. Med Sci Sports Exerc 2007; 39: 665-671
18. Kelley MA, Laufe MD, Millman RP, Peterson DD. Ventilatory response to hypercapnia before and after athletic training. Respir Physiol 1984; 55: 393-400
19. Kilen A, Larsson TH, Jørgensen M, Johansen L, Jørgensen S, Nordsborg NB. Effects of 12 weeks high-intensity & reduced-volume training in elite athletes. PloS one 2014; 9: e95025
20. Lucía A, Hoyos J, Pérez M, Chicharro JL. Heart rate and performance parameters in elite cyclists: a longitudinal study. Med Sci Sports Exerc 2000; 32: 1777-1782
21. Menz V, Strobl J, Faulhaber M, Gatterer H, Burtscher M. Effect of 3-week high-intensity interval training on VO2max, total haemoglobin mass, plasma and blood volume in well-trained athletes. Eur J Appl Physiol 2015; 115: 2349-2356
22. Milsom WK, Abe AS, Andradeb DV, Tattersall GJ. Evolutionary trends in airway CO 2/H+ chemoreception. Respir Physiol Neurobiol 2004; 144: 191-202
23. Miyamura M, Ishida K. Adaptive changes in hypercapnic ventilatory response during training and detraining. Eur J Appl Physiol Occup Physiol 1990; 60: 353-359
24. Myers J, Gademan M, Brunner K, Kottman W, Boesch C, Dubach P. Effects of high-intensity training on indices of ventilatory efficiency in chronic heart failure. J Cardiopulm Rehabil Prev 2012; 32: 9-16
25. Ohkuwa T, Fujitsuka N, Utsuno T, Miyamura M. Ventilatory response to hypercapnia in sprint and long-distance swimmers. Eur J Appl Physiol Occup Physiol 1980; 43: 235-241
26. Richardson RS, Verstraete D, Johnson S, Luetkemeier M, Stray-Gundersen J. Evidence of a secondary hypervolemia in trained man following acute high intensity exercise. Int J Sports Med 1996; 17: 243-247
27. Salazar-Martínez E, Gatterer H, Burtscher M, Naranjo Orellana J, Santalla A. Influence of inspiratory muscle training on ventilatory efficiency and cycling performance in normorxia and hypoxia. Front Physiol 2017; 8: 133
28. Salazar-Martínez E, Terrados N, Burtscher M, Santalla A, Orellana JN. Ventilatory efficiency and breathing pattern in world-class cyclists: A three-year observational study. Respir Physiol Neurobiol 2016; 229: 17-23
29. Viaño-Santasmarinas J, Rey E, Carballeira S, Padrón-Cabo A. Effects of High-Intensity Interval Training with Different Interval Durations on Physical Performance in Handball Players. The Journal of Strength & Conditioning Research 2017, DOI:
30. Wahl P, Mathes S, Achtzehn S, Bloch W, Mester J. Active vs. passive recovery during high-intensity training influences hormonal response. Int J Sports Med 2014; 35: 583-589
31. Williams BM, Kraemer RR. Comparison of cardiorespiratory and metabolic responses in kettlebell high-intensity interval training versus sprint interval cycling. J Strength Cond Res 2015; 29: 3317-3325

Title of tables

Table 1: Baseline characteristics of the HIITG and the CG (n=16).

Table 2: Training data (training performed during the 3-week training period) for HIITG and CG.

Table 3: Ventilatory and performance variables analyzed during the intervention period in both groups.

Title of figures

Figure 1: Relationship between ventilatory efficiency (V_E/VCO_2 slope) and maximum oxygen uptake (VO_{2max}) with data from both groups and both test.

Figure 2: Relationship between ventilatory efficiency (V_E/VCO_2 slope) and the time in 400m test (T400m) with data from both groups and both test.

Table 1: Baseline age and physical characteristics of the training and the control group.

	HIT (n=8)		CG (n=8)
Age (years)	25.6±3.2		25±3.4
Height (cm)	181.6±5.8		178.7±4.9
Weight (kg)	74.2±5.5		75.2±6.3
VO_{2max} ($ml \cdot kg \cdot min^{-1}$)	68.4±2.7		67±6.5
Data are presented as mean ± SD			

Table 2: Training data (all endurance training performed during the 3-week training period) for HITG and CG.

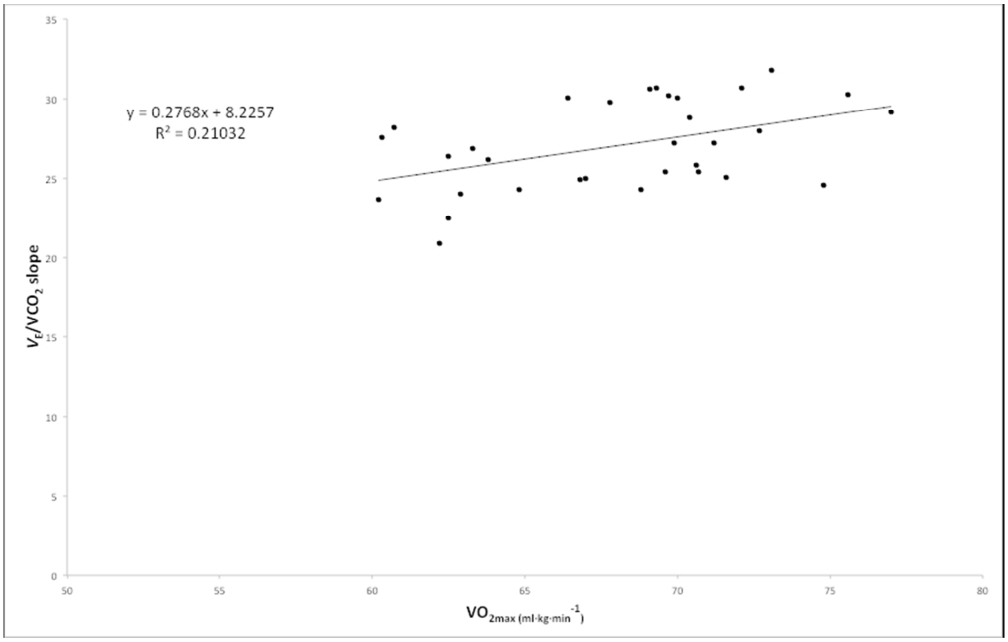
	HIT group (n=8)				CG (n=8)		
	Endurance (min)	Borg	Trimp		Endurance (min)	Borg	Trimp
Week 1	329.1±134.5	13.7±1.9	3570.1±1914.8		331.2±95.8	13.2±0.9	4431.8±1549.3
Week 2	248±90.4	14.1±1.7	3163.5±1320.6		272.5±196.2	14.1±2.8	3522.2±2718.2
Week 3	297.5±123.2	14.5±1.3	4062.1±1632.3		390.7±192.7	14.8±4.1	5129.7±2439.8
Total	874.6±212.8	14.1±1.1	13,098.8±4734.5		994.5±418.1	14±2.3	12,326.7±5706

Data are presented as mean ± SD
TRIMP training impulse (perceived exertion × endurance training session time)

Table 3: Ventilatory and performance variables analyzed before (Pre) and after (Post) the intervention period in both groups.

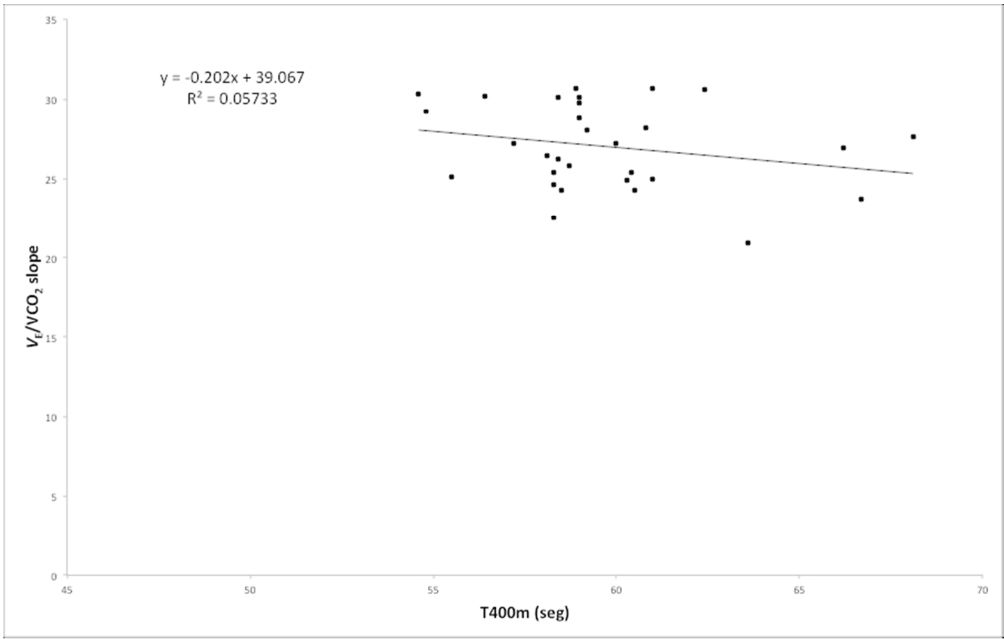
	HIT group (n=8)		Effect Size (Cohen´s d)	CG (n=8)		Effect Size (Cohen´s d)	ANOVA (interaction) time x group
	Pre	Post		Pre	Post		
V_E/VCO_2 slope (up to VT_2)	28.1±2.2	27.4±2.3	0.24	26.1±3.2	26.5±3.1	0.21	0.522
V_E/VCO_2 slope (up to exhaustion)	30.8±2.7	30.5±3.3	0.11	30.2±3.4	30.4±3.8	0.07	0.757
V_{Emax} (l·min^{-1})	181.8±17.8	184±19.4	0.38	181.7±14.5	181.5±17.5	0.02	0.607
VO_{2max} (ml·kg·min^{-1})	68.4±2.7	69.8±1.8	0.43	67.1±6.5	66.8±5.7	0.05	0.246
T400m (seg)	59.6±1.6	58.3±1.9	0.75	60.8±4.6	60.2±3.9	0.25	0.557

V_E/VCO_2 slope, ventilatory efficiency; V_{Emax}, maximum ventilation; VO_{2max}, maximum oxygen uptake; T400m, time in 400m field test
* p<0.05
<0.2 small, 0.5 medium and >0.8 large effect size (ES)



Relationship between ventilatory efficiency (VE/VCO2 slope) and maximum oxygen uptake (VO2max) with data from both groups and both test.

114x72mm (200 x 200 DPI)



Relationship between ventilatory efficiency (VE/VCO2 slope) and the time in 400m test (T400m) with data from both groups and both test.

114x72mm (200 x 200 DPI)

Biology of Sport

Ventilatory efficiency response is unaffected by fitness level, ergometer settings, age or body mass index in male-athletes.

Type:

Original paper

Abstract:

Objectives
The aim of this study was to evaluate the ventilatory efficiency (VE/VCO2 slope) and the respiratory control (Vt/Ti slope) in a wide range of athletes and describe the influence of fitness level, age, ergometer or BMI in these parameters.

Material and methods
Ninety-one males (30.4±10.53 years; 175.52±7.45 cm; 71.99±9.35 kg) were analysed retrospectively for the study.

Results
Ventilatory efficiency reacted similarly in athletes independently of the fitness level, the age, the BMI or the ergometer used for testing. No significant differences were found in VE/VCO2 slope and the Vt/Ti slope between variables analyzed ($P>0.05$). The slope of the predictive equations was similar in all cases studied in VE/VCO2 slope and the Vt/Ti slope. Moreover, the central control impulse of respiration was neither affected by the variables studied.

Conclusions
These observations suggest that ventilatory efficiency (VE/VCO2 slope) could be a variable fixed by the respiratory system which tends to response similarly in athletes.

Keywords:

respiratory, Body Composition, exercise testing, VE/VCO2 slope, Efficiency

VENTILATORY EFFICIENCY RESPONSE IS UNAFFECTED BY FITNESS LEVEL, ERGOMETER SETTINGS, AGE OR BODY MASS INDEX IN MALE-ATHLETES

Head Title

VENTILATORY EFFICIENCY IN ATHLETES

Abstract

The aim of this study was to evaluate the ventilatory efficiency (V_E/VCO_2 slope) and the respiratory control (Vt/Ti slope) in a wide range of athletes and describe the influence of fitness level, age, ergometer or BMI in these parameters. Ninety-one males (30.4±10.53 years; 175.52±7.45 cm; 71.99±9.35 kg) were analysed retrospectively for the study. Ventilatory efficiency reacted similarly in athletes independently of the fitness level, the age, the BMI or the ergometer used for testing. No significant differences were found in V_E/VCO_2 slope and the Vt/Ti slope between variables analyzed ($P>0.05$). The slope of the predictive equations was similar in all cases studied in V_E/VCO_2 slope and the Vt/Ti slope. Moreover, the central control impulse of respiration was neither affected by the variables studied. These observations suggest that ventilatory efficiency (V_E/VCO_2 slope) could be a variable fixed by the respiratory system which tends to response similarly in athletes.

Key words: V_E/VCO_2 slope / Respiratory / Exercise Testing / Body composition / Efficiency

1. Introduction

Ventilatory efficiency can be defined as the relationship between carbon dioxide production (VCO_2) and ventilation (V_E) during an incremental exercise test (1). It has been reported several ways for measuring ventilator efficiency (2, 3). However, using the slope of the relationship between VCO_2 and V_E (V_E/ VCO_2 slope) has been suggested as the best way for achieving a correct evaluation of the ventilatory efficiency during an incremental exercise test (4). It adds information about the global ventilatory efficiency throughout entire test and not only at one metabolic rate as it happens with the equivalent of CO_2 (V_E/VCO_2) (5).

Ventilatory efficiency has been widely studied in patients suffering congestive heart failure (CHF) or cardio-respiratory weakness (6-9). Values exceeding 34 are considered abnormal (1, 10) or indicative of the inefficiency of the respiratory system (2). In healthy subjects, it has been reported an inter-variability in the values of the V_E/VCO_2 slope (from 19 to 32) (3).

The role and importance of ventilatory efficiency in human sport performance remains controversial. The matching of ventilation and perfusion in the lungs is the primary determinant of ventilatory efficiency (REF-101 VOL). Conditions where the CO_2 production is elevated, as exercise, seem to play an essential role in the ventilatory control (11). In this regard, it could be possible that a greater efficiency of CO_2 elimination during exercise might allow a higher sport performance. However, in elite-juvenile cyclists, no relationship has been found between maximal oxygen uptake (VO_{2max}) and V_E/VCO_2 slope (2). In the same way, it has been reported that changes in sport performance in world class-cyclists over three competitive seasons are not related to changes in V_E/VCO_2 slope (5). In syinchornonized swimmers, ventilatory efficiency kept unalterable by working conditions during the apneic episodes (12). Data from our research group revealed that submaximal cycling performance was not related to ventilatory efficiency response (13). We hypothesized that increments in CO_2 production are linked to proportional increment in ventilation in spite of a fitness level.

Physiologic dead space (V_D/V_T) has been suggested as a variable that could modify ventilatory efficiency response (3). Age and anthropometric characteristics might influence V_D/V_T (14). However, in children, ventilatory efficiency was not affected by sex despite differences in anthropometric characteristics (15). Same results were found in adults, no age or sex differences were found on ventilatory efficiency in healthy participants (3). However, from our knowledge there are not studies which evaluate ventilatory efficiency in athletes with different characteristics. Thus, measuring the influence of age and BMI on ventilatory efficiency is necessary in order to better clarify if there are differences between athletes with different characteristics.

Regarding type of ergometer, a test dependency has been reported in healthy woman, but not in males (16). The authors explained these results due to a small amount of arterial hypoxemia coupled with a small amount of arterial hypercapnia in woman (16). However, from our knowledge this is the only study mainly focused in this analysis. Thus, further evaluation in athletes is necessary in order to evaluate the influence of type of ergometer on ventilatory efficiency response.

Although, ventilatory efficiency has been already studied in healthy people, this variable has not been wide studied in athletes. Contrary to ventilatory efficiency, breathing pattern has been wide studied in athletes (17-19). V_E can be decomposed into the product of two components: (a) central inspiratory activity, known as "driving" and expressed as the relationship between Vt and inspiratory time (Vt/Ti) and (b) the inspiration-expiration alternation, known as "timing", and expressed by the relationship between Ti and the total duration of the breathing cycle (Ti/Ttot) (20, 21). Vt/Ti and Ti/Ttot responses during incremental exercise appear to be stable and independent of fitness level (12, 17). Studying the relationship between V_E, Vt/Ti and VCO_2 we could know if the central control of respiration makes ventilatory efficiency (V_E/VCO_2 slope) behaves similar in athletes independently of their characteristics.

Thus, the aim of this study was to evaluate ventilatory efficiency and respiratory control in a wide range of athletes and describe the influence of fitness level, age, ergometer or BMI in these parameters. In this regard, we hypothesize that ventilatory efficiency could be an inborn characteristic which responses similar in athletes independently of fitness level, age, ergometer or BMI.

2. Materials and methods

Subjects

From a large amount of incremental exercise tests carried out in our laboratory, we selected those which were carried out by healthy sporty people from different endurance sport disciplines (running, cycling, triathlon) and fitness level (amateur, semi-professional). Ninety-one active and healthy males (30.4±10.53 years; 175.52±7.45 cm; 71.99±9.35 kg) were analysed retrospectively for the study. Participants were classified in different groups depending on ergometer used for testing, BMI, age and VO_{2max} (Treadmill (n=37); Cycle ergometer (n=54); BMI: 18-25 (n=70); 25-30 (n=21); Age: 16-25 (n=40); 25-35 (n=16); 35-45 (n=23); >45 (n=12); VO_{2max}: <45VO_{2max} (37.8±7.4 $ml \cdot kg^{-1} \cdot min^{-1}$; n=43); >45 VO_{2max} 51.9±5.1 $ml \cdot kg^{-1} \cdot min^{-1}$; (n=48)). Fitness level classification was set according Paap and Takken (22) proposal. Cardio-respiratory variables are shown in Table 1.

Participants were tested in our laboratory for different previous proposes. All previous studies were approved by the ethical committee of Pablo Olavide University and conformed to standards of treatment of human participants in research as outlined in the Fifth Declaration of Helsinki. Participants were informed (both in writing and orally) about all testing and training procedures and gave their written informed consent to participate prior to entering the study.

Procedures

From the tests carried out in our laboratory we selected those performed with the same protocol on cycle ergometer (Ergoselek 200, Ergoline, Germany) or on treadmill (Ergorun 8, Down electronics, Germany). Each participant performed a maximum incremental exercise tests with gas analysis. During each test, oxygen uptake (VO_2), carbon dioxide output (VCO_2), respiratory exchange ratio (RER), ventilation (V_E), breathing frequency (f_R), tidal volume (VT), oxygen equivalent ($EqVO_2$), carbon dioxide equivalent ($EqCO_2$), driving (Vt/Ti) and timing (Ti/Ttot) were recorded every 5 seconds breath by breath with a gas analyser (Cpx última, medical graphics, USA). The system was calibrated prior to each test with gas mixtures of known concentration. After 4 min of warming up, participants started the test at 50W and then the load was increased by 25W each minute until volitional exhaustion on cycle ergometer. On treadmill, after 4 min of warming up the participants started the test at 7 km/h and the velocity was increased by 1 km/h each minute until volitional exhaustion. Tests were carried out under similar and controlled environmental conditions (20-25°C; 45-55% relative humidity). Achievement of maximal oxygen uptake (VO_{2max}) was accepted when a plateau was found in the relationship between VO_2 and power output or when three of the four criteria for maximal VO_{2max} were obtained (23).

Ventilatory efficiency and breathing pattern

The ventilatory efficiency of each subject was calculated from the slope of the relationship between VCO_2 and V_E during each test. To exclude the influence due to respiratory compensation for acidosis during highly intensive exercise, the V_E/VCO_2 slope was determined from the beginning of the test until the second ventilatory threshold (VT_2). VT_2 was identified using the criteria of increase in both ventilatory equivalents - EqO_2 and $EqCO_2$ - and end tidal partial pressure of oxygen ($PETO_2$) with no concomitant increase in end tidal partial pressure of carbon dioxide ($PETCO_2$) or decrease in $PETCO_2$ (24, 25). The value of the slope representing the relationship between V_E and Vt/Ti during each test (Vt/Ti slope) was used to test the central component of respiration.

Statistical analysis

Data are expressed as mean ± SD and with Cohen's d effect size (ES) for each variable. Subjects were included in different groups depend on fitness level, ergometer used for testing, BMI and age. The normal distribution of the data in each group was checked by means of the Shapiro–Wilk test. The homogeneity of variance was evaluated by Levene´s test. To compare the mean values obtained of V_E/VCO_2 slope and Vt/Ti slope in each group the following statistical tests were carried out. Student's T-Test for independent samples was used to compare fitness level groups and type of ergometer groups. Kruskal–Wallis H-test was carried out to compare mean values between BMI groups. One-way ANOVA test were used to compare mean values between age groups. The Bonferroni test was selected as a post hoc test. Linear regression analysis was performed for each group between V_E (dependent variable) and VCO_2 (independent variable) and Vt/Ti (dependent variable) with data from each subject. Effect sizes (ES) were also calculated using Cohen´s d. The level of significance was set at $P<0.05$ for each statistical analysis. An ES of $d<0.2$ was considered small, 0.5 medium and $d>0.8$ large (26).

3. Results

Data on the ventilatory efficiency and ventilatory control evaluation are shown in Table 2. The statistical analysis found non-significant differences ($P>0.05$) both for the V_E/VCO_2 slope and Vt/Ti slope for all the variables included in the analysis (ergometer, BMI, age, and fitness level). Effect size analysis showed a low ES between cycle-ergometer and treadmill testing on V_E/VCO_2 slope and Vt/Ti slope (0.29 and 0.09 respectively). Regarding BMI, a low-medium ES was found between groups in V_E/VCO_2 slope and Vt/Ti slope (0.46 and 0.24 respectively).

No age effect was found in V_E/VCO_2 slope and in Vt/Ti slope (0.16 and 0.15 respectively). Fitness level showed a low ES for diffrences between groups in V_E/VCO_2 slope (0.33) and Vt/Ti slope (0.33). Table 3 shows the predicting equations for V_E/VCO_2 slope after regression and statistical analysis. The slope of the predictive equations was similar in all cases studied (Table 3). Figure 1 shows the regression lines for each variable studied.

4. Discussion

To the best of our knowledge, this is the first study which evaluates the influence of ergometer, age, BMI and fitness level on ventilatory efficiency in athletes. We hypothesized that ventilatory efficiency could behave independently of before mentioned variables in athletes. The main finding of this study was that ventilatory efficiency is not influenced by the ergometer used for testing, the athlete´s age, BMI or fitness level. These findings support the hypothesis that ventilatory efficiency could be an inborn characteristic which react independently of fitness level, anthropometric profile, age or the ergometer used for testing.

Ventilatory efficiency has been proposed as an effectiveness method to detect cardiorespiratory weakness and healthy problems (6, 7, 9). Values exceeding 34 are indicative of the inefficiency of the cardiorespiratory system (1, 27). However, it is not as clear that athletes with better ventilatory efficiency are those who perform a high sport performance. In our study, no differences were found in V_E/VCO_2 slope between athletes with a low VO_{2max} *vs* high VO_{2max} (23.4±4.2 and 24.8±4.1, respectively). The slope of the predictive equations was also similar in both cases (24.12 and 24.88, respectively) (Table 3) (Figure 1). Similar mean values of efficiency were found in world-class cyclists over 3-year period (24.6±3.1; 23.6±2.7; 24.8±2.6) (5). Even though, these cyclists were tested with a totally different protocol (50W each 4 min), gas analyzer and they had a higher VO_{2max} (77.5±6.2 $ml \cdot kg^{-1} \cdot min^{-1}$), they showed similar values of ventilatory efficiency to our subjects. In this way, changes in sport performance (peak power output) were no related to changes in V_E/VCO_2 slope or

VO_{2max} in world-class cyclists (5). In juvenile cyclists, no relationship was found between VO_{2max} and V_E/VCO_2 slope (2). No correlation was found between V_E/VCO_2 slope and VO_{2max} in sport students before and after inspiratory muscle training neither normoxia nor hypoxia (13). Thus, our results and the evidence reported before, help us to confirm the hypothesis that V_E/VCO_2 slope could not be a variable related to sport performance. In this regard, it has been suggested that if an athlete has poor cardio-respiratory efficiency (high V_E/VCO_2 slope) it has no bearing on their maximal ability to use oxygen (2) or achieving a high performance (5). Therefore, V_E/VCO_2 slope has not efficacy in quantifying the performance of the physiological systems which support an athlete's ability to perform at high oxygen uptakes (2).

In terms of age and BMI, controversial data about ventilatory efficiency has been reported. On the one hand, Sun, Hansen (3) carried out an evaluation of ventilatory efficiency on healthy people without significant difference between sexes and ages. On the other hand, ventilatory efficiency showed a sex and age dependence in healthy subjects (4). In children, ventilatory efficiency response was not affected by sex (15). In our study, we could not compare ventilatory efficiency between sexes due to the small sample size in females. Regarding age analysis, no differences were found between age groups in V_E/VCO_2 slope (Table 2). These results are in concordance with previous studies (3, 15). Physiologic dead space (V_D/V_T) has been proposed as a variable that could modify ventilatory efficiency in healthy subjects (3). Maturation and age could modify the V_D/V_T (14) and as a consequence ventilatory efficiency. In our subjects, the mean values obtained in age groups were similar to values measured in children (15) (Table 2). Thus, ventilatory efficiency might be a variable no affected by age or anthropometric characteristics in healthy athletes.

With reference to type of ergometer, we did not find difference between subjects tested on treadmill or cycle ergometer on ventilatory efficiency response (Table 2). We compared world-class cyclist ventilatory efficiency data (5), who were tested with a different gas analyzer and with a different protocol (50W/4 min), with our subjects (25W/min). The mean values obtained were similar in both cases (~24). Same results were obtained in men but not in women, suggesting an independence of test mode evaluation (16) and an independence of speed used in the test on ventilatory efficiency response (28). In the first study (16), the protocol used (4 min of walking at 72 $m \cdot min^{-1}$ and 0% grade. At the end of minutes 4, 7 and 10, the speed was increased by 10 $m \cdot min^{-1}$) was totally different to ours. In the second (28), they did not found different between the fast (25W/min) and the slow protocol (five work rate increments of equal size each 4 min). But one more time, the slope values reported (24.19 and 23.23 respectively) were in concordance with our results. In the same way, Sun, Hansen (3) found an absence of the effect of laboratory site or ergometer in ventilatory efficiency evaluation, with a greater reproducibility for V_E/VCO_2 slope (online data supplement). The slope of the predictive equations was similar in all cases studied (Table 3). Accordingly to these results, type of ergometer or protocol used might not modify the ventilatory efficiency response in healthy athletes.

Furthermore ventilatory efficiency analysis, we carried out an analysis of driving component of respiration (Vt/Ti slope). As it occurs with V_E/VCO_2 slope, the increment in the driving impulse was similar in all our subjects and it was independent of age, fitness level, BMI or ergometer (Table 2). In all these cases, the increment in driving impulse was closely to ~40. This indicates that the increases in V_E during a progressive exercise are associated with a proportional increase in the inspiratory driving activity without any alteration in the relationship between inspiration and expiration, even at the highest working intensities (Figure 3) (5). Thus, the lineal relationship of V_E with Vt/Ti and VCO_2 suggest that the main factor conditioning the stability of ventilatory efficiency (as V_E/VCO_2 slope) could be the central impulse of respiration (Vt/Ti).

Some limitations have to be addressed. First, this study was retrospectively and we could not measure body composition variables in our subjects. Further investigations taking into account body composition variables would be necessary in order to better clarify if body composition could influence ventilatory efficiency response. Lastly, we could not include females in our study due to the low sample size. New research to evaluate the influence of gender on ventilatory efficiency is necessary in order to better clarify the involvement of this variable on ventilatory efficiency response.

Based on the previous evidence reported and in our results, we propose a nomogram for assessing ventilatory efficiency (V_E/VCO_2 slope) (Figure 2). This nomogram might help to carry out a better evaluation of ventilatory efficiency in athletes completing the proposal of Naranjo, Centeno (12). In addition, they could help to detect easily cardio-respiratory problems or deficiencies in respiration control when an incremental test is carried out in athletes.

In summary, ventilatory efficiency reacted similarly in athletes independently of the fitness level, the age, the BMI or the ergometer used for testing. Moreover, the central control impulse of respiration was neither affected by the variables studied (Figure 3). These observations suggest that ventilatory efficiency (V_E/VCO_2 slope) could be a variable fixed by the respiratory system which tends to response similarly in athletes. Finally, ventilatory efficiency could be assessed easily during an incremental test in athletes using the nomogram proposed.

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Conflict of interest

No potential conflict of interest was reported by the authors.

References

1. Arena R, Myers J, Hsu L, Peberdy MA, Pinkstaff S, Bensimhon D, et al. The minute ventilation/carbon dioxide production slope is prognostically superior to the oxygen uptake efficiency slope. J Card Fail. 2007;13(6):462-9.
2. Brown SJ, Raman A, Schlader Z, Stannard SR. Ventilatory efficiency in juvenile elite cyclists. Journal of Science and Medicine in Sport. 2013;16(3):266-70.
3. Sun X-G, Hansen JE, Garatachea N, Storer TW, Wasserman K. Ventilatory efficiency during exercise in healthy subjects. American journal of respiratory and critical care medicine. 2002;166(11):1443-8.
4. Habedank D, Reindl I, Vietzke G, Bauer U, Sperfeld A, Gläser S, et al. Ventilatory efficiency and exercise tolerance in 101 healthy volunteers. European journal of applied physiology and occupational physiology. 1998;77(5):421-6.
5. Salazar-Martínez E, Terrados N, Burtscher M, Santalla A, Naranjo Orellana J. Ventilatory efficiency and breathing pattern in world-class cyclists: A three-year observational study. Respiratory Physiology & Neurobiology. 2016;229:17-23.
6. Arena R, Myers J, Guazzi M. The clinical and research applications of aerobic capacity and ventilatory efficiency in heart failure: an evidence-based review. Heart Fail Rev. 2008;13(2):245-69.
7. Ingle L, Goode K, Carroll S, Sloan R, Boyes C, Cleland JG, et al. Prognostic value of the VE/VCO2 slope calculated from different time intervals in patients with suspected heart failure. Int J Cardiol. 2007;118(3):350-5.
8. Laveneziana P, Agostoni P, Mignatti A, Mushtaq S, Colombo P, Sims D, et al. Effect of Acute β-blocker Withholding on Ventilatory Efficiency in Patients With Advanced Chronic Heart Failure. Journal of cardiac failure. 2010;16(7):548-55.
9. Magrì D, Limongelli G, Re F, Agostoni P, Zachara E, Correale M, et al. Cardiopulmonary exercise test and sudden cardiac death risk in hypertrophic cardiomyopathy. Heart. 2016:heartjnl-2015-308453.
10. Arena R, Guazzi M, Myers J. Prognostic value of end-tidal carbon dioxide during exercise testing in heart failure. Int J Cardiol. 2007;117(1):103-8.
11. Milsom WK, Abe AS, Andradeb DV, Tattersall GJ. Evolutionary trends in airway CO 2/H+ chemoreception. Respiratory physiology & neurobiology. 2004;144(2):191-202.
12. Naranjo J, Centeno RA, Carranza MD, Cayetano M. A test for evaluation of exercise with apneic episodes in synchronized swimming. International journal of sports medicine. 2006;27(12):1000-4.
13. Salazar-Martínez E, Gatterer H, Burtscher M, Naranjo Orellana J, Santalla A. Influence of inspiratory muscle training on ventilatory efficiency and cycling performance in normoxia and hypoxia. Frontiers in Physiology. 2017;8:133.
14. Mummery HJ, Stolp BW, deL Dear G, Doar PO, Natoli MJ, Boso AE, et al. Effects of age and exercise on physiological dead space during simulated dives at 2.8 ATA. Journal of Applied Physiology. 2003;94(2):507-17.
15. Guerrero L, Naranjo J, Carranza MD. Influence of gender on ventilatory efficiency during exercise in young children. J Sports Sci. 2008;26(13):1455-7.
16. Davis JA, Tyminski TA, Soriano AC, Dorado S, Costello KB, Sorrentino KM, et al. Exercise test mode dependency for ventilatory efficiency in women but not men. Clinical physiology and functional imaging. 2006;26(2):72-8.
17. Lucia A, Carvajal A, Calderon FJ, Alfonso A, Chicharro JL. Breathing pattern in highly competitive cyclists during incremental exercise. Eur J Appl Physiol Occup Physiol. 1999;79(6):512-21.
18. Lucia A, Hoyos J, Pardo J, Chicharro JL. Effects of endurance training on the breathing pattern of professional cyclists. Jpn J Physiol. 2001;51(2):133-41.

19. Scheuermann BW, Kowalchuk JM. Breathing patterns during slow and fast ramp exercise in man. Exp Physiol. 1999;84(1):109-20.
20. Milic-Emili J. Recent advances in clinical assessment of control of breathing. Lung. 1982;160(1):1-17.
21. Milic-Emili J, Grunstein MM. Drive and timing components of ventilation. Chest. 1976;70(1 Suppl):131-3.
22. Paap D, Takken T. Reference values for cardiopulmonary exercise testing in healthy adults: a systematic review. Expert review of cardiovascular therapy. 2014;12(12):1439-53.
23. Howley ET, Bassett DR, Welch HG. Criteria for maximal oxygen uptake: review and commentary. Medicine and science in sports and exercise. 1995;27(9):1292-301.
24. Lucía A, Hoyos J, Pérez M, Chicharro JL. Heart rate and performance parameters in elite cyclists: a longitudinal study. Medicine and Science in Sports and Exercise. 2000;32(10):1777-82.
25. Skinner JS, McLellan TH. The transition from aerobic to anaerobic metabolism. Research quarterly for exercise and sport. 1980;51(1):234-48.
26. Cohen J. Statistical power analysis. 1988.
27. Chase P, Arena R, Myers J, Abella J, Peberdy MA, Guazzi M, et al. Relation of the prognostic value of ventilatory efficiency to body mass index in patients with heart failure. The American journal of cardiology. 2008;101(3):348-52.
28. Davis JA, Sorrentino KM, Soriano AC, Pham PH, Dorado S. Is ventilatory efficiency dependent on the speed of the exercise test protocol in healthy men and women? Clinical physiology and functional imaging. 2006;26(2):67-71.

Tables

Table 1: Maximum cardio-respiratory values during the incremental exercise test (n=91)

	VO_2 (ml·min^{-1})	VCO_2 (ml·min^{-1})	f_R (br·min^{-1})	VT (ml)	V_E (l·min^{-1})	Ti/Ttot	Vt/Ti (ml·sec^{-1})	$PETCO_2$ (mmHg)
Mean	3219.8	4051.9	51.2	2240.4	112.8	0.41	4823.1	43.6
SD	571.1	808.2	11.6	424.6	26.2	0.05	962.6	6.6

SD, standard deviation; VO_2, oxygen uptake; VCO_2, carbon dioxide output; f_R, breathing frequency; Vt, tidal volume; V_E, ventilation; Ti/Ttot, timing; Vt/Ti, driving; $PETCO_2$, end tidal pressure of carbon dioxide.

1

Table 2: Comparison of Mean±SD values of the V_E/VCO_2 slope and Vt/Ti slope for the treadmill and cycleergometer cardiopulmonary exercise tests, the body mass index (BMI) ranges (18-25; 25-30), age ranges (16-25; 25-35; 35-45; >45) and fitness level (<45 VO_2max; >45 VO_2max) in athletes.

	ERGOMETER				BMI ($kg \cdot m^{-2}$)				AGE (years)							FITNESS LEVEL: VO_2max ($ml \cdot kg^{-1} \cdot min^{-1}$)			
	Cycle (n=37)	Treadmill (n=54)	p-value	Effect Size	18-25 (n=70)	25-30 (n=21)	p-value	Effect Size	16-25 (n=40)	25-35 (n=16)	35-45 (n=23)	>45 (n=12)	p-value	Effect Size	<45 VO_2max (n=43)	>45 VO_2max (n=48)	p-value	Effect Size
V_E/VCO_2 slope	23.6±3.8	24.8±4.4	0.146	0.29	24.5±4.1	22.6±4	0.067	0.46	24.3±3.8	22.9±4.5	24.1±4.6	25.6±3.7	0.146	0.16	23.4±4.2	24.8±4.1	0.111	0.33
Vt/Ti slope	38.7±6.5	39.4±6.3	0.592	0.09	38.8±6.3	40.4±7.1	0.336	0.26	38.8±6.4	38.4±6.2	40.7±6.6	38.1±6.7	0.416	0.15	40.5±6.3	38.3±6.3	0.100	0.33

*Significantly different between groups ($p < 0.05$).
[§] Large effect size (ES≥0.8).

Table 3: Predictive equations for the ventilatory efficiency response.

	Predictive equations					
	a	b	r^2	r	Standard Error	p-value
Ergometer						
Cycle	25.81	0.964	0.929	0.964	0.07	0.000*
Treadmill	24.11	0.913	0.834	0.913	0.106	0.000*
BMI ($kg \cdot m^{-2}$)						
18-25	24.70	0.948	0.899	0.948	0.064	0.000*
25-30	24.66	0.950	0.903	0.950	0.125	0.000*
AGE (years)						
16-25 (n=48)	24.91	0.963	0.927	0.963	0.072	0.000*
25-35 (n=28)	24.49	0.936	0.875	0.936	0.137	0.000*
35-45 (n=23)	23.28	0.890	0.793	0.890	0.177	0.000*
>45 (n=12)	26.03	0.986	0.973	0.986	0.088	0.000*
FITNESS LEVEL: VO_{2max} ($ml \cdot kg^{-1} \cdot min^{-1}$)						
<45 VO_{2max} (n=43)	24.12	0.945	0.893	0.945	0.094	0.000*
>45 VO_{2max} (n=62)	24.88	0.941	0.885	0.941	0.081	0.000*

* Level of significance ($p < 0.05$).

$y = a \cdot x + b$ ($y = V_E$ (ventilation); $x = VCO_2$ (carbon dioxide output); $a = V_E/VCO_2$ slope; b= y-intercept)

Figure legends

Figure 1: Evaluation of ventilatory efficiency (V_E/VCO_2 slope) showing regression lines measured in each group (Treadmill (n=37); Cycle ergometer (n=54); BMI: 18-25 (n=70); 25-30 (n=21); Age: 16-25 (n=40); 25-35 (n=16); 35-45 (n=23); >45 (n=12); VO_{2max}: <45VO_{2max} (n=43); >45 VO_{2max} (n=48)). All groups showed a similar linear adjustment.

Figure 2: Graph showing the linear relation between carbon dioxide output (VCO_2) and ventilation (V_E) with data from all sample size (n=91). This can be used as a nomogram for assessing ventilatory efficiency in healthy athletes during exercise regardless of the ergometer, fitness level, age or body mass index.

Figure 3: Graph showing the linear relation between and ventilation (V_E) and driving impulse (Vt/Ti) with data from all sample size (n=91). Central impulse of respiration responded similar in all participants regardless of the ergometer, fitness level, age or body mass index.

Table 1: Maximum cardio-respiratory values during the incremental exercise test (n=91)

	VO_2 (ml·min^{-1})	VCO_2 (ml·min^{-1})	f_R (br·min^{-1})	VT (ml)	V_E (l·min^{-1})	Ti/Ttot	Vt/Ti (ml·sec^{-1})	$PETCO_2$ (mmHg)
Mean	3219.8	4051.9	51.2	2240.4	112.8	0.41	4823.1	43.6
SD	571.1	808.2	11.6	424.6	26.2	0.05	962.6	6.6

SD, standard deviation; VO_2, oxygen uptake; VCO_2, carbon dioxide output; f_R, breathing frequency; Vt, tidal volume; V_E, ventilation; Ti/Ttot, timing; Vt/Ti, driving; $PETCO_2$, end tidal pressure of carbon dioxide.

Table 2
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Table 2: Comparison of Mean±SD values of the V_E/VCO_2 slope and Vt/Ti slope for the treadmill and cycleergometer cardiopulmonary exercise tests, the body mass index (BMI) ranges (18-25; 25-30), age ranges (16-25; 25-35; 35-45; >45) and fitness level (<45 VO_2max; >45 VO_2max) in athletes.

	ERGOMETER				BMI ($kg \cdot m^{-2}$)				AGE (years)						FITNESS LEVEL: VO_2max ($ml \cdot kg^{-1} \cdot min^{-1}$)			
	Cycle (n=37)	Treadmill (n=54)	p-value	Effect Size	18-25 (n=70)	25-30 (n=21)	p-value	Effect Size	16-25 (n=40)	25-35 (n=16)	35-45 (n=23)	>45 (n=12)	p-value	Effect Size	<45 VO_2max (n=43)	>45 VO_2max (n=48)	p-value	Effect Size
V_E/VCO_2 slope	23.6±3.8	24.8±4.4	0.146	0.29	24.5±4.1	22.6±4	0.067	0.46	24.3±3.8	22.9±4.5	24.1±4.6	25.6±3.7	0.146	0.16	23.4±4.2	24.8±4.1	0.111	0.33
Vt/Ti slope	38.7±6.5	39.4±6.3	0.592	0.09	38.8±6.3	40.4±7.1	0.336	0.26	38.8±6.4	38.4±6.2	40.7±6.6	38.1±6.7	0.416	0.15	40.5±6.3	38.3±6.3	0.100	0.33

*Significantly different between groups ($p < 0.05$).
§ Large effect size (ES≥0.8).

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Table 3: Predictive equations for the ventilatory efficiency response.

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Figure 1

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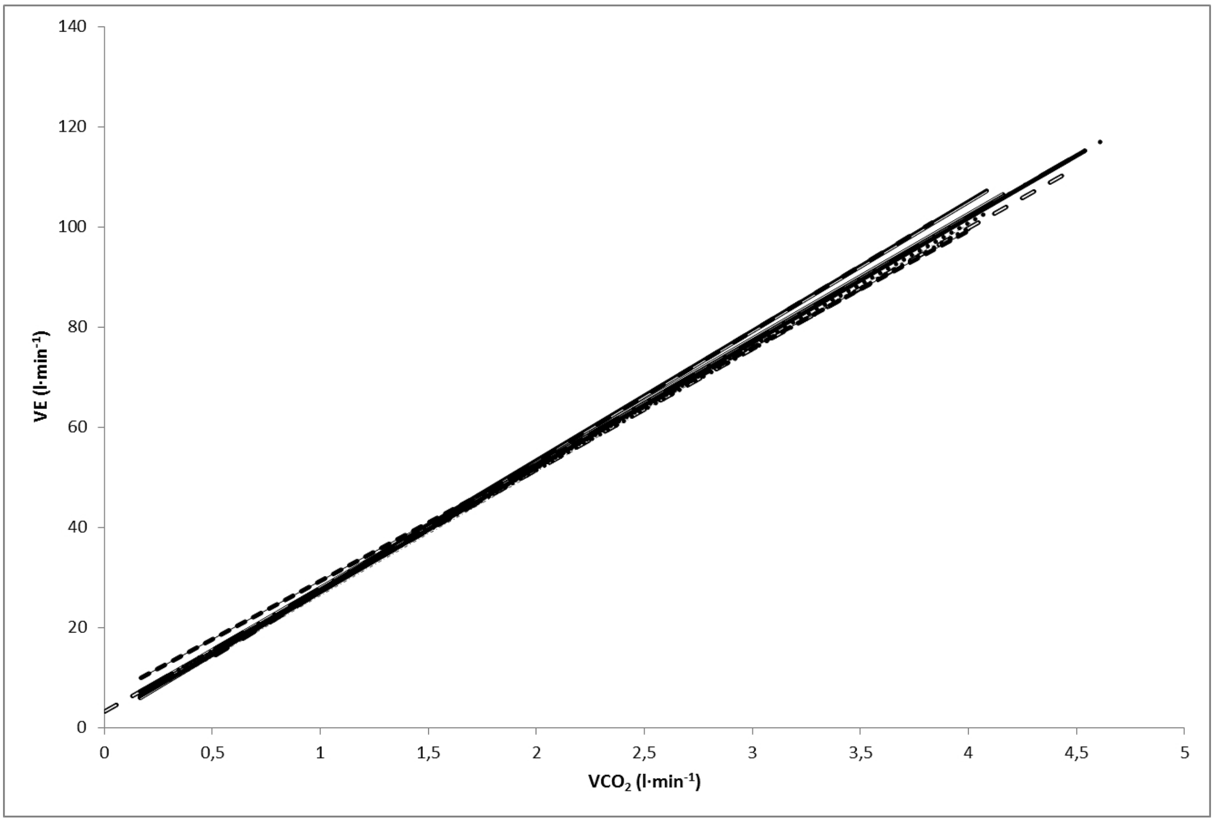
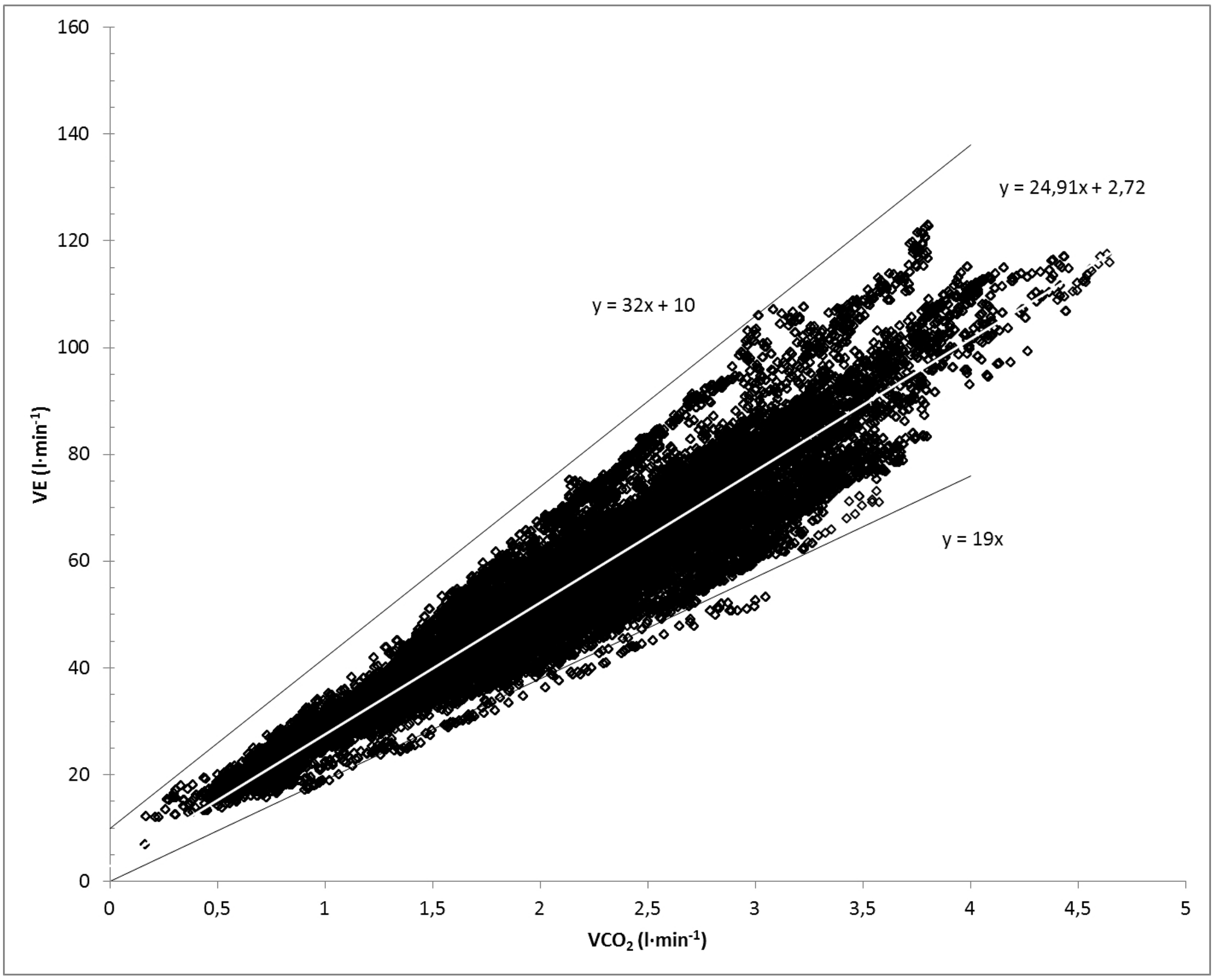


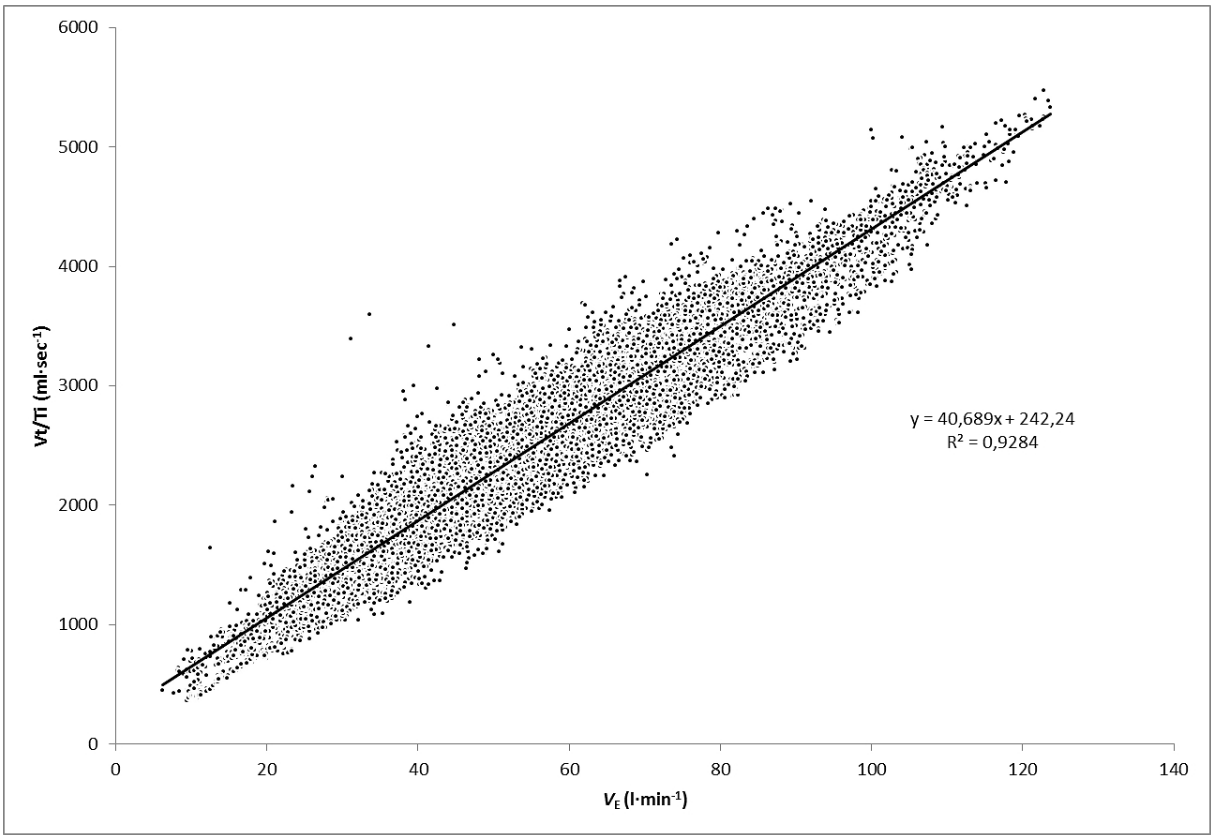
Evaluation of ventilatory efficiency (VE/VCO2 slope) showing regression lines measured in each group (Treadmill (n=37); Cycle ergometer (n=54); BMI: 18-25 (n=70); 25-30 (n=21); Age: 16-25 (n=40); 25-35 (n=16); 35-45 (n=23); >45 (n=12); VO2max: <45VO2max (n=43); >45 VO2max (n=48)). All groups showed a similar linear adjustment.

Figure 2

Biology of Sport



Graph showing the linear relation between carbon dioxide output (VCO2) and ventilation (VE) with data from all sample size (n=91). This can be used as a nomogram for assessing ventilatory efficiency in healthy athletes during exercise regardless of the ergometer, fitness level, age or body mass index.



Graph showing the linear relation between and ventilation (VE) and driving impulse (Vt/Ti) with data from all sample size (n=91). Central impulse of respiration responded similar in all participants regardless of the ergometer, fitness level, age or body mass index.

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Manuscript body 1 - Download source file (92.62 kB)

Tables

Table 1 - Download source file (15.2 kB)
Table 1: Maximum cardio-respiratory values during the incremental exercise test (n=91)

Table 2 - Download source file (16.57 kB)
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Table 3 - Download source file (16.6 kB)
Table 3: Predictive equations for the ventilatory efficiency response.

Figures

Figure 1 - Download source file (53.18 kB)
Evaluation of ventilatory efficiency (VE/VCO2 slope) showing regression lines measured in each group (Treadmill (n=37); Cycle ergometer (n=54); BMI: 18-25 (n=70); 25-30 (n=21); Age: 16-25 (n=40); 25-35 (n=16); 35-45 (n=23); >45 (n=12); VO2max: <45VO2max (n=43); >45 VO2max (n=48)). All groups showed a similar linear adjustment.

Figure 2 - Download source file (132.26 kB)
Graph showing the linear relation between carbon dioxide output (VCO2) and ventilation (VE) with data from all sample size (n=91). This can be used as a nomogram for assessing ventilatory efficiency in healthy athletes during exercise regardless of the ergometer, fitness level, age or body mass index.

Figure 3 - Download source file (174.34 kB)
Graph showing the linear relation between and ventilation (VE) and driving impulse (Vt/Ti) with data from all sample size (n=91). Central impulse of respiration responded similar in all participants regardless of the ergometer, fitness level, age or body mass index.